FOOD AND DRUG ADMINISTRATION
National Center for Toxicological Research (NCTR)

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National Center for Toxicological Research (NCTR)
Science Advisory Board (SAB)

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P R O C E E D I N G S (8:00 a.m.)

Agenda Item: Welcome and Overview

DR. ASCHNER: Thank you, everybody. I know we are a little bit challenged these days, so I appreciate all of your efforts. Everybody is obviously timely. We have a busy agenda, and if you want to see the agenda, it's in the pod to your right.

I'll give a couple of introductory remarks, basically just to let you know the purpose of this, and we'll go and introduce the members of the Scientific Advisory Board, and then you can see the schedule. I'll give Bill an opportunity to talk about the state of the Center. And most of today is going to be presentations by the different FDA centers, and towards the afternoon we're going to hear from a couple of the divisions, which we review on a yearly basis.

We'll finish with the other divisions tomorrow, and that's going to be followed Tuesday afternoon and Wednesday by review by subcommittee of one of the divisions.

Again, welcome. What I'd like to do first is just acknowledge the members of the advisory board. I'm Michael Aschner, if you don't know me. I'm at the Albert Einstein College of Medicine, and my interest is in neurotoxicology. As mentioned already, the designated

federal officer is Donna Mendrick, and she's at Silver Spring in Maryland at the National Center for Toxicological Research, and special thanks go to Donna. We were obviously supposed to be all together today in Little Rock, but everyone knows the reason we're doing everything remotely. It's a challenge when we're together, and it's a challenge when we're just Zooming in. But I can tell you, the NCTR is close to heart to Donna and we appreciate everything that she has done for the center.

The advisory members are, I'll go alphabetically, Mary Ellen Cosenza, she's interested in regulatory toxicology, and she's the president of MEC Regulatory and Toxicology Consulting in Los Angeles.

Patty Ganey, who's a professor at Michigan State University in the department of pharmacology and toxicology, and her interest is in liver toxicology.

Charles Kaspar, who is a professor and chair of the department of bacteriology at the University of Wisconsin.

Greg Lanza, whose expertise in biomedical molecular imaging, and he's a professor of medicine in the cardiovascular division at Washington University in St. Louis.

Ken Ramos, who is an expert in genetics, toxicological genetics, and he's the executive director of

the Texas A&M Institute of Bioscience and Technology, Texas A&M University.

John-Michael Sauer, who is interested in drug metabolism and toxicology in general, and is the consumer representative.

The next one is Steve Stice. His interest is in environmental science and stem cell biology. He is the D.W. Brooks Distinguished Professor and Georgia Research Alliance Eminent Scholar in the Center for Regenerative Bioscience at the University of Georgia.

Last but not least is Dr. Alexander Tropsha, who is the K.H. Lee Distinguished Professor, associate dean, for pharmacoinformatics and data science at the University of North Carolina in Chapel Hill.

These are the SAB members. I don't think we're going to go around the table this year. It's a little bit complex, and introduce all the members, all the representatives from the different FDA centers. But we'll be able to hear from you a little bit later as you give your individual talks.

With that, that's all I want to say for the introduction. As I said, we're going to have a day-and-a-half of a busy schedule, and you have opportunities to ask questions at the end of the talks, SAB members, as already noted.

Without further ado, I'd like to again thank Donna. I don't know if you want to say anything else, Donna, or whether I should go directly and ask Bill to show his presentations.

Thank you, Donna, and I'll give you the floor.

Agenda Item: Conflict of Interest Statement and "Housekeeping Items"

DR. MENDRICK: Thank you so much, Miki, and thank you for the kind words.

Good morning, everyone. I am Donna Mendrick, the Designated Federal Official, and I would like to welcome everyone to the NCTR Science Advisory Board meeting. We appreciate the time and diligent work of our board members in preparing for this meeting and for the forthcoming deliberations. I and the board wish to thank the FDA regulatory centers and NIEHS for their participation in this meeting, and my NCTR colleagues for all their efforts preparing for this meeting.

Let me say a word about my role as the Designated Federal Official. As the DFO for this meeting, I serve as the liaison between the board and the agency. I'm responsible for ensuring all provisions of the Federal Advisory Committee Act, FACA, are met regarding the operations of the SAB. Also, in my role as DFO for the board, a critical responsibility is to work with

appropriate agency officials to ensure that all appropriate ethics regulations are satisfied. In that capacity, board members are briefed on the provisions of the federal conflict of interest laws, and in addition, each participant has filed a standard government financial disclosure report.

Regarding the meeting operations, we have a full agenda yet strive to ensure adequate time for thorough deliberations. This special note for all presenters, board members, and the participants; please keep your video off and mute your phone until you speak. Announce your name when you do so, as this meeting is being recorded and a transcript will be posted on our website. Again, please turn off your video and mute your phone after you're finished.

Pursuant to the fact that we had a public comment period scheduled for today, however, no one expressed an interest, so we'll just continue with the meeting. We won't have that hour set aside.

I would like to add that during presentations and discussion, if board members require greater clarification on an issue regarding participation with attendees or the audience, they may request this information during the meeting to the chair or myself. In accordance with FACA, minutes of this meeting will be prepared as will a

transcript. Both will be posted to our external website. Thus, remember this is a public meeting.

In closing, I wish to thank the board for their participation in today's meeting. Thanks, everyone.

Bill Slikker is up next.

Agenda item: State of the Center

DR. SLIKKER: All right, good morning everyone. This is Bill Slikker. I'm the director of NCTR. I'm so glad that you're all here today. I want to thank you for making the effort to be here, both the Scientific Advisory Board members as well as our representatives from various centers and ORA. Really, it's a pleasure to have the opportunity to learn more about the NCTR. Many of you have had a chance to visit. As Donna mentioned, I wish that we could be together today, I could go around and shake your hands and talk with you before the meeting started, but we'll have to go with this format based on a very small organism that's disrupting a fabulous group meeting of our SAB. But I think we can make it all work by using our slides and our communication tools that we have available, so I want to thank Donna in particular for rising to the occasion. Also Michael and Justin and many, many others who have worked on getting the technical aspects of this worked out.

Again, a special thanks, of course, to our SAB members who are here to listen to our presentations, give us guidance for the future, as well as the representatives from the various centers.

So I'll go ahead and begin my presentation just by mentioning that the NCTR, as we found out at the last SAB, all the centers are special, all the centers are unique, but certainly NCTR is one of those centers that has that unique quality to it, as well.

One thing I want to point out is that we do have a large facility. It's somewhere around 500 acres of space that belongs entirely to FDA, and it also has somewhere around 1.2 million square feet of floor space for scientific achievement. We also share this campus with the Arkansas lab of ORA, very happy that they've been on board now for more than 18 years, and also many segments of other parts of FDA are on this campus, as well. But the unique thing about our campus is that since it belongs to FDA, we can modify it to fit for purpose.

With that, I just want to say something about the organization. You're going to hear from all the division directors in those boxes at the bottom, those six boxes at the bottom. So I'm not going to spend a lot of time introducing them, except that they, that's the business end

of our organization. The research activities are the key, the driver of our organization.

However, I must say that this middle row of individuals here and offices is really critical to making all that happen, and that is possible because we have some really quality people leading those various groups, including Tucker Patterson, who's involved in all parts of research at NCTR, including the work with the concepts and proposals that go through review, coordinating all that activity, as well as a manuscript review process.

The office of scientific coordination, Dr. Schnackenberg, Brad does a great job in running that organization that supports all the research activity on our campus. The Office of Research is where Tucker and others reside to handle all the proposals and the research activities within NCTR.

The regulatory compliance and risk management group -- very important, because this is our safety group, biosafety for the entire center. Well-managed by Raj Nayak, and we really appreciate Dr. Nayak's leadership of that group. Office of Management, run by Winona Cason, who is our executive officer, and she does a fabulous job there along with our CFO, Charlotte, and those two really manage the process along with their team to make sure that we have

the best kind of planning and the best kind of funding opportunities for NCTR.

Finally, the last box on the right, the associate director for regulatory activities is Donna, and I've already mentioned how critical she is as our main force out on the White Oak campus to keep us connected, and also of course to manage our Scientific Advisory Board meetings.

So thank you again, Donna, as we move on to our next slide.

This gives you an idea of the number of staff, and this varies from time to time. As you'll see, most of these slides are from a few months back, as far as the datasets are concerned, but you can see that we do have around 300 government employees. And most of these are researchers. We're really very lean on management, it's somewhere around 15 percent or less. And we also have contractors, which do a lot of the work having to do with animal husbandry, maintenance of the facilities, all the cleaning aspects, the pathology group, and the security -- all very critical and we work very closely with them to get the job done, and they're an excellent group to be working with.

And then also the ORISE, which our postdoctoral fellowship program. It's a training program, we're very proud of the ability to train folks on campus, always

looking for new individuals that may want to join in the training program, and also the idea that we feel very good about placing these individuals in great positions as they move forward in their careers.

Let me just say that the goal can be really divided into three major areas. The first one being very, very important, and that is to advance the science and the knowledge about all kinds of FDA-regulated products, and this is to support both personal care as well as animal and human health. This is a broad area, but the idea is for us to generate data that can serve as a basis for FDA decision-making, and we do that in close collaboration, which is the second goal, with the other centers. We do that because most of our protocols are actually in the collaboration with other centers, because we need to know the needs of the regulatory aspects of FDA, and we need to use that information to devise protocols and projects that will support and provide the needed data for FDA decision-making.

And then finally on the right is the interactions globally. This is very critical because, as you know, America imports many different foodstuffs, and also various ingredients for medicines, and many other items including devices from abroad. We need to be able to make sure that

we have good connections with those individuals from abroad.

Moving on, what we're looking at here is the interaction with the other centers and the staff at NCTR, and what you see is the division of many different groups of activities, including the various product line centers, as well as universities and other government institutions. This means that this collaborations helps us make sure we're focused on the FDA goals and reach group kinds of datasets that can be useful to the agency.

You all are pretty much aware that NCTR has a tremendous portfolio of activities going on, fundamental expertise, and this fundamental expertise really covers many different areas, which are all listed here. I'm not going to go into detail here, but you're going to be hearing about this directly from the division directors over the next small amount of time.

The next slide is really looking more toward the emerging technology, so another activity that occupies about half of the resources of NCTR is developing these new emerging technologies, and then moving toward validating them for use for FDA decision-making. And that's a critical feature of what we do. And we also have advisory functions which we do with either global organizations or organizations within America.

The science at the NCTR is really quite diverse. We not only of course develop and evaluate new kinds of technologies to help FDA make better decisions, but we also look at resolving issues that may be of regulatory concern, as well as, as I mentioned, advancing those toolsets that can be useful for the future. We also have been focusing recently, in the last six months, of course, on COVID-19, and with this is over 10 different studies that have been initiated to attack the issue of COVID-19, not only on the vaccine development aspect but also on therapeutics that may be used to treat COVID-19. And then also a whole host of other studies that have been developed, many of them with the COVID focus, and other focuses important to FDA.

So over some 50 new protocols have been developed just in the last several months to focus on this COVID-19 issue. These have been done in conjunction mainly with the other centers of FDA.

That brings us to the whole idea of bridging the toxicology paradigms, and this is where you really use those newer technologies that have been developed throughout the world, and some right here at NCTR, and put them through their paces so we can make sure that they're applicable to FDA issues and concerns, and can be used routinely by FDA. So that particular approach I want to exemplify on this slide, and the idea is that this is a

challenge, because you're moving oftentimes from animal models or in vitro tests, with human cells, but you need to get that information to be valuable to the intact human or animal, for safety concerns. So that's what we spend our time doing, is making sure that we can link that new approach, the in vitro approach, the cell culture approach, back to whole animal data and clinical studies, so that we can make sure they're useful to FDA decision-making.

One of those examples can be shown here with the Center for Tobacco Product-NCTR Inhalation Core Facility, where we're able to generate data from in vivo --this is the rodents species, and the idea is that we can use this data to extrapolate to the human situation. But in addition to that, it also can be very useful to have a way to look at this in vitro. So using this human air-liquid interface model, one can start to generate data with human cells in culture, and understand and collaborate and work between the whole-animal work and the human cells in culture to make sure that we're on target to generate data that's useful to the FDA.

In this way, you can look at a variety of endpoints, well-controlled in vitro, where we can look at not only the structure of the actual tissue, the lung tissue, as well as the various cell types that are there. You can also look at the activity of the cilia which are so

important to the normal function of the respiratory tract, and you can identify and look at the health of goblet cells, basal cells, also analyze for tight junctions, et cetera, using this particular approach.

And the idea then, of course, is that you can use this then to look at new issues, not only having to do with the Center for Tobacco Product type issues that have to do with tobacco agents, but also by working with other groups as well, to look at extrapolation of pharmacokinetics within the lung system. So this can be really useful, for example here, when you have developed a new protocol that can examine the effects of coronavirus antiviral drugs and use this approach then for screening and for drug repositioning, and you can see here that this project is in concert, in collaboration, with CBER, as well as CDER, and this is critical for us to make sure that we're on target by generating data that is useful for the FDA decisionmakers.

Let me just move on then to many other in vitro approaches that we have developed over the last 10 or 15 years. This one has to do with drug-induced cardiomyopathy, but it really has to do with the idea of developing an in vitro system which then you can analyze using your omics technology. Certainly, Bill Mattes will go over this in more detail.

Also, the use of patient-specific induced stem cells. These can be either agents that may have cancer activities, and you can evaluate those in the same sort of in vitro fashion, always comparing back to the in vivo data where you can, with animals or in human, with human data.

We have also expanded this more recently now to look at an in vitro testicular model so you can look at reproductive function using in vitro approaches, and just to give you some further examples, we've also developed models that are useful to study the blood-brain barrier in vitro, as well as nervous system cells in vitro, and a variety of other in vitro approaches that can be useful.

Let me just move on then to the microbiology area. This is an important division that was, it will be undergoing further review, as all of our divisions do. It's time for its review about every five or six years, and it will be coming up on Wednesday afternoon and Thursday of this week. But this group has been amazing leaders in the field of microbiology, and actually was studying the microbiome before it was well-described in that term. And also it hosts our area where we have the folks that study viruses. We're very glad that over 10, 12 years ago, the very SAB that you're sitting on suggested that we add virology to our microbiology group. We did that, and we were so glad that we did because now those investigators

are leading many of the studies to study COVID-19, for example.

Just one example of that is a project there by one of our virologists, and you'll be hearing more about this later, but the whole idea is to develop a special tool that can be useful to study the coronaviruses in general and COVID-19 in particular. These approaches then are available for even level 2 laboratories. They can be used there without having to go to a level 3 lab to get pertinent data.

I want to finish up my brief review of some of the division activity by looking at the bioinformatics/biostatistics area. This is one of our newer divisions, it's only about seven years old now, but Weida Tong and group have really taken us to a new level by devising various strategies, interact specifically with the FDA centers. Certainly here we can see a lot of interaction going on with the Center for Drug Evaluation and Research with some of their platforms that have been developed hand-in-hand between NCTR and CDER. DASH, for example, and some of the other tools that have been developed.

So this approach of working with the other centers is very much alive and well within the bioinformatics and biostatistical area. One of those

examples is the FDALabel approach which has been around now for five or six years, but this particular survey approach allows one to look at thousands and thousands of FDA labels and get to them quickly, analyze the outcome, and use it to help develop new safety processes.

Recently there was a survey done, and it was very successful, over 200 CDER users per month of this particular software package that was made available by NCTR, working with CDER. We also find that there's many within CBER, the Center for Biologics, that also use this approach. It had a very high satisfaction rate of over 88 percent of those that have responded to date. So we're very proud of these kinds of individual efforts that allow our center to help other centers develop datasets and processes that are useful to them.

And the same thing goes for these databases that are mentioned here. We've already talked about the FDA label, but there are many other ones, including the Endocrine Disruptor Knowledge Base, there's also those that deal with DILI liver dysfunction, et cetera. So these databases and approaches are again so useful to the other centers to allow them to quickly analyze new data coming in, put it in a study, and allow them to make faster, more efficient decisions.

One of those examples is a new protocol, again, focused on treating COVID-19. This one, as you can see, has collaborators in CDER, from CDRH. The idea here is that if one could find the best way to focus your attention on various possible drugs they could use to treat COVID-19, then you'd want to know the sort of priority rankings of which one you want to look at first. And this particular approach allows you to do that.

So let me just switch then to some individual areas that we're very proud of at the NCTR. The NanoCore, of course, has amazing array of equipment. It is built up between NCTR and ORA, it shares campus here, Jefferson labs, with us. It is a resource that is available to all of the parts of FDA, and many have collaborated with us to use this equipment to help identify issues with products that are in the nanomaterial range.

Also we have an amazing in vivo imaging capability that we use for preclinical and nonclinical study, and we have two different MRIs, including now a large bore machine as well as two different PETs connected to CT. These instruments are available throughout the agency, and we appreciate those collaborative projects that we've been able to generate under the leadership of Serguei Liachenko and Sherry Ferguson and others within the

Division of Neurotoxicology. You'll be hearing more about that later.

Let me just finish up my introduction of NCTR and talk a little bit about one of our projects we began really through additional funding that came in in the 2019 budget. We were very fortunate to get some funding so we began the FDA Perinatal Health Center of Excellence. This really includes a broad area of development, everything from the maternal exposures to lactation, also premature infants, neonates, as well as development through childhood. This particular opportunity is available to all the different offices and centers within the FDA.

The beneficial part of this, of course, is that there's very little information usually available on maternal-fetal pairs, and they represent a unique kind of responsibility at FDA. Also, as you know, there's many preterm and term infants and neonates that need special support, and oftentimes drugs are not readily studied in these populations, so it's one area of focus that we feel is necessary throughout the FDA, and we can help move that area forward.

We've been doing that through a number of steps. The first one was to get the funding in 2019, then to develop leadership groups within the FDA with representatives from each one of the centers and ORA, so

that they could have input into the process to evaluate proposals that come in to this funding source and to pick the very best ones to move forward with support. We did that, and the first year we had some 14 that were funded.

More recently, last year, for example, we had smaller amounts of support available that particular year, but were able to evaluate 10 proposals and select three of those for funding. So right now we have a total of 17 ongoing projects. We're very fortunate to have funding available again in 2021, and therefore, we are right now in the process of reviewing up to 14 or so new proposals that will be evaluated by this liaison group between the various centers and ORA to select those that require and would deserve funding, and we'll move those forward and fund those in the very first part of -- well, actually end of this year, and be available for all of next year's support.

With that, I just want to mention a couple other items that are important to the NCTR and the FDA. We have our leadership role within the National Toxicology Program for all of FDA, and this is connected, of course, to NIH through NIEHS, to CDC to NIOSH, and NCTR has a representative, Goncalo, representative for all of FDA at the table.

This group has been around for over 25 years, and has resulted in funding and many projects at the NCTR and

in conjunction with the other centers and NIEHS and NIOSH in some cases, to get critical work done. Some of these areas, of course, have been fostered by tight interaction between NIEHS and their Division of Toxicology, as well as the work at all the centers and NCTR.

You can see here a list of some of the agents that have been evaluated over the last 25-plus years, high-profile agents and ones where we needed information for the entire United States, but particularly important for FDA. This information was generated so that good, firm decisions can be made based on the reports and many publications, well over 200, that have come out showing off this important work to the world, so that everybody could benefit from this particular use of resources.

Let me just finish up by mentioning our global outreach, now in its tenth year. We'll be celebrating our tenth year here very soon. This global outreach is really a consortium, you might say, a special coalition of regulatory groups from around the world. It includes representatives from Japan, from now one of new members, Switzerland, Swissmedic, from Brazil. One of our really large memberships, which is the EU, including the EMA, EFSA, and JRC, which represents over 25 countries. We have the Republic of Korea, we have New Zealand, Australia, China, as well as Argentina, Canada, and Singapore. This

group is really quite active, and one of its main roles is not only to survey and see where we can work together and collaborate and where data needs are, but also to sponsor a global summit. The Global Summit on Regulatory Science is now entering its tenth year.

Here we see the results from meetings that were held annually. This one in 2018 was on risk and benefit of dietary supplements and herbal medicines in the era of data science. This was held in Beijing. And the 2019 one was held and cosponsored by the JRC in northern Italy and really focused on nanotechnology and nanoplastics. So we're moving ahead now with our meeting for 2020, which will be our tenth anniversary meeting, and it's focusing on emerging technologies and their application to regulatory science.

We're really proud of the lineup of some 70 speakers we have for this meeting, including the director of NIH, the director of EFSA, and the commissioner of the FDA, as well as our chief scientist and many, many more. We'll have plenary speakers to talk about the general overview, and then we'll have in-depth scientific presentations representing speakers from 14 different countries all around the world to focus on this very important era of emerging technologies and how we can best apply them to make regulatory decisions.

This will be held virtually, as you might guess, but it will be held in a time zone of September 29 through 30, with a live chat and question and answer period on September 30, that it would be great for all of you to join, if you have time.

So let me just stop there and say that our mission, of course, is to develop datasets for FDA decision-making and develop new tools and approaches that can be used to generate data for FDA needs. I just want to thank all the individuals that you'll be hearing from for the rest of the day and then tomorrow and the next day. Our staff and our scientists are excellent, and I really appreciate them preparing their presentations for you to see, and for your input and questions and good advice that we want to have from you to move us into the future. We really have this Scientific Advisory Board to generate interest and generate new ideas that we can look at in the future, and for over 25 years now this Science Advisory Board group for NCTR has been doing such, and so we appreciate your efforts to do the same.

Thank you all very much, and I'd be happy to answer any questions you may have.

DR. ASCHNER: This is Miki Aschner. Thank you very much, Bill, for the presentation. I have essentially one question, and it's not directly what you talked about.

I just wanted to get some sense on how this pandemic actually might have affected the projects. I was happy to see that you are working on issues that are related to COVID-19, but in general I also recognize the last Scientific Advisory Board meeting was only about eight months ago. I'm just wondering how the NCTR was generally impacted by this pandemic, and what do you see coming up in the next few months?

DR. SLIKKER: Really good question. Thanks to Tucker Patterson riding herd over the new proposals and concepts that have been coming in, we actually have a large number, some 50 new proposals, of which 29 or such are focused on COVID, but other really important items, as well. So that really is an uptick in the number of proposals that have been written during the last six months.

Also, for manuscripts, we have a record bumper crop of manuscripts. Well over 100 in the last six months have been written and put through the review process, which is quite strenuous, before they can be submitted to a journal. And all that work has been really, in some ways, enhanced by a lot of telework that's had to occur. But certainly people have been, the scientists have been very productive and the support staff and making sure that we

move these products forward, both in terms of new concepts and proposals, as well as manuscripts.

The other thing, of course, is that many of these projects have now been started, and this is in part because of funding that's been made available to us through a variety of means, some directly from FDA, and due to some of the supplements that have come in from Congress, so that's helped move this forward. But also just the sheer energy that's been expended.

And also, because of our contractors in animal care and pathology and our essential staff, we have been able to maintain many of our projects, especially those involving long-term studies, so that we can continue to generate data. Now, as we look at the opportunity for more individuals to return to campus over a period of time, more of these projects will be started up and completed. So we're very pleased with how we are handling this by keeping many people who can on telework kinds of approaches to limit the number of people actually on campus, makes it safer, and of course we follow all the guidelines and rules from CDC and FDA to make sure that we keep our staff safe. That's the most important thing.

But we're able to do that by having many people telework but still be very, very productive. So thanks for that question. Any other questions from you or others?

DR. ASCHNER: I don't see any other questions. Thank you very much, Bill. Thank you for leading the NCTR through good times and difficult times, as we are experiencing now. I think we all acknowledge the work that you're doing. So thank you and everybody else who's working at the NCTR.

(Housekeeping remarks.)

Agenda item: Subcommittee Review of Division of Neurotoxicology

DR. ASCHNER: The next item on the agenda is the NCTR Scientific Advisory Board subcommittee visit. This was held December 4 and 5 last year, after the regular meeting, the review of the NCTR. We were tasked with reviewing the Division of Neurotoxicology. We had two members on the committee, myself and Dr. Greg Lanza, and we had several experts who participated, giving their expertise on the subject matter. These were Drs. Pat McConville, and Dr. Ed Levin, and Dr. Wei Zhang.

I'll start with some general comments for the division as a whole, and then I'll move on and talk specifically about each of the three different research areas that were identified in the Division of Neurotoxicology.

The way that we structured our report was basically, we had five different sections to each of them:

integration into the FDA mission, the quality of the research, new technologies and approaches that we thought should be considered at this time, areas that are relevant or less relevant to the FDA's public health mission, and then finally, recommendations.

I'm not going to go through everything because we don't have enough time, but hopefully, as I go through the division as a whole and to the different research areas, you'll get a good sense and we'll hear later the response from the Neurotoxicology Division as to where they are and where they should aim to be in the future.

I'll start with the overall comments for the Neurotoxicology Division. Overall, we were very impressed with the leadership of the division, and we commend Dr. Ferguson who took the reins of this division about three years ago, and she definitely continues in the foundation that was laid by her predecessor, Drs. Bill Slikker and Merle Paule.

We thought that the work in the division was very well aligned with the FDA mission, and that the work had major translational value for regulatory decisions. One of the things that was most apparent to the subcommittee and especially for those of us who have been on this committee before, came off and are back on, is the multiple and ongoing collaborations with other FDA centers and that's

something I think we've heard from Bill already and hopefully this will continue, because I think it's very informative to the division and it helps the other centers in terms of the regulatory decisions and moving the science forward.

We thought that there was a lot of internal and external collaborations, as I mentioned. These were important for the NCTR, and we encourage the NCTR to continue to pursue those as much as possible when you are advancing your future research programs.

Some of our recommendations for the division as a whole. We thought that the DNT continues to perform an outstanding service to the NCTR, the FDA, and neurotoxicology, in general, with the research programs that were outlined to us, not just in testing toxicant exposure effects on various endpoints. There was a lot of emphasis on their behavior. But we also thought that it helps to develop better understanding of key processes and whether it's neuro behavior or other functions that are disrupted or sensitive to various toxicants.

Their strategy provides immediate answers concerning the individual drugs and chemicals under study, and, most importantly, builds on a foundation of understanding and facilitating neurotoxicology investigation going forward.

One of the things is that we recommend is that DNT continue to integrate neurotoxicological investigation across levels of analysis from in silico, in vitro -- we've heard about some of these again from Bill -- perform studies in vertebrates in different rodent models, and all the way to nonhuman primates. We think that's important, allowing complementary and alternative models, has obviously advantages and disadvantages, but in general, especially when you're testing hypothesis-driven questions, it can accelerate the research.

It's important that DNT have leeway to balance between proving service studies for specific toxicity questions for the FDA, we think this is important, but also keep balance in answering fundamental studies to advance the field of neurotoxicology and facilitate future investigations. I'll talk about it a little bit later, we heard a lot about some new programs on various brain disorders, Alzheimer's, Parkinson's disease, ADHD, TBI. We think these are important studies to pursue. We thought that a lot of the work that was described to us, especially as it relates to the blood-brain barrier, is unique, and the focus of the NCTR on the blood-brain barrier may be very useful in assessing new drugs that might be effective in combating these diseases.

One thing that we thought there could be some improvement is we got the general sense that it's basically leadership that determines what kind of programs are going to move forward, and we thought that it would be beneficial to make use of the subcommittee members or the NCTR in general, the DNT specifically, can come up with some method to decide exactly how they might want to review protocols in the future. I think having, or we think, that having some folks from the outside, as I said, either from the subcommittee or from other institutions, is going to be helpful in strengthening and making priorities for the DNT and NCTR in general.

The final thing for the division as a whole, we did have some question about a couple of the programs. We really maybe wasn't explained very well to us, but we were questioning the relevance of these programs. One that was specifically noted was regarding tobacco regulation. We felt that the research was very hypothesis-drive, but we didn't get the sense that it was important in impacting various regulatory decisions by the FDA.

The first division that we reviewed was the imaging in neurotoxicology. This division is led by Drs. Liachenko and Zhang. We felt that the quality of the research in this division was overall outstanding. It had potential to impact the NCTR in a very significant and

unambiguous way, and we further concurred that the research fit very well with the mission of the FDA and that the technologies were helpful in facilitating FDA approval and review of the safety and efficacy of novel agents and toxicants.

As I mentioned, the quality was deemed to be very impressive. The division basically is divided into two different subdivisions, if you wish. One of them is the PET-CT and the other one is the MRI. As far as the MRI, which is led by Dr. Liachenko, we thought that it was particularly mature and diverse. We've heard many presentations where this kind of instrumentation and expertise was leveraged into various projects and had very impactful effect on these projects.

We noted few areas that included the T-based imaging neurotoxicology, which are deemed very important to the mission of NCTR as a whole, and these included, we thought the strides that have been made over the last few years are actually tremendous. These are related to pediatric anesthesia, where there's been a lot of imaging, gadolinium contrast agents, iron nanoparticles, and some of the research on nicotine and tobacco.

As far as the PET-CT, again, we commend the progress that's been done in this subdivision of DNT imaging. Most of the focus has been recently on apoptosis

imaging, which we think is very important, it fits very well with anesthesia studies that are being conducted in rats and nonhuman primates. We thought that the quality of the imaging data was very high, and that Dr. Zhang has done a great job, actually given some of the limitations that you have onsite in getting various labeling compounds, fluorine-18 labeled compounds that are necessary for this kind of imaging.

As far as the opportunity or new technologies that should be considered, we recognized a couple of those. We thought that you should consider expanding the image analysis and access to analysis expertise and hardware and software. As I mentioned before, the imaging core has become relatively mature, but we felt that there might be -- this is the time to try to move it to the next step, get some more standard analysis software and hardware that will help you in storing image reconstruction, backup, archiving, visualization, and quantification of the data.

For the PET, the biodistribution imaging, we thought it would not take very large effort and it wouldn't be too expensive to expand on these PET imaging applications to include longer-lived isotope-based labeling, and we generally felt that these are going to be helpful in terms of developing the AD, the TD, the TBI, studies that are under way in the Neurotoxicology Division.

We thought that these tracers would have great potential and should actually provide you with much more meaningful data when you're doing those studies. You can look specifically at various tau proteins, alpha-synuclein, there's imaging methodology that can be used which would be helpful in those studies.

The last, I think for this division, we commend the interaction between this core, the imaging core, and the group that's working on the blood-brain barrier, which I'll discuss later. Again, we think that there might be some additional studies where you might wish to incorporate imaging biomarkers to the blood-brain barrier studies that are being conducted, which would provide a new and valuable platform for the NCTR in terms of understanding more of the molecular mechanisms that are associated with some of these diseases.

The major recommendations for this division are basically keep doing what you're doing. Try to get some additional equipment. We're always there for DNT and NCTR in general. Hopefully, you can secure additional resources. We think that you are on the right track, this division specifically, or subdivision. And we think that by adding those issues, those items that I've mentioned, you will be able to expand on your imaging capability and

work and integrate it to some of the other programs that are ongoing within the Division of Neurotoxicology.

The second research area is the core on Alzheimer's disease, Parkinson's disease, and TBI, and there's a number of researchers that are involved, Drs. Sarkar and Imam and Dr. Rosas-Hernandez. Again, we thought that this research area fits very well in the mission of the DNT. We thought that over the years this division, or this group, has led, has significant accomplishments in a relatively short period of time, and again, we thought the areas of research are relevant to the mission of the Division of Neurotoxicology.

As far as the quality of the research, strength and opportunities, the quality of this core, the science is of high quality in many aspects. First of all, the core has made some novel discoveries. Those include serum exosomes as a potential biomarker for Alzheimer's disease, increased blood-brain barrier permeability as a condition of cerebral amyloid angiopathy.

Secondly, the core has developed critical in vivo and in vitro models. They are using a number of transgenic models for studies in Alzheimer's disease. They've developed some transgenic rat models, in vitro biaxial stretch model for TBI, and microelectrode arrays for screening of neurotoxicants and microphysiological systems

for blood-brain barrier modeling. And finally, as I mentioned before, in collaboration with the imaging center, some of these techniques have been integrated including studies on lipodome and peripheral biomarkers of Parkinson's disease, using 3D, MALDI, MS, and select scan Lipidyzer.

We think the division has state-of-the-art techniques. The studies are very broad, yet very focused, and generally are of great translational value. A couple opportunities that should be considered are, again, using the MRI CT facility maybe to a greater extent, and we do recognize there are limitations in terms of personnel, but there are some methodologies that certainly could be incorporated in terms of imaging and looking at the blood-brain barrier dysfunction.

Overall recommendations for this core is basically to keep doing the work. I'm not going to go through all of them. I've mentioned some of this as I was mumbling about the research that's going on; primarily I think we came to a conclusion that it would be beneficial to add some, or even enhance, the MRI and the CT parts of the studies on these various neurological disorders.

The last division that we reviewed is the development neurotoxicology core research area. This is led by Dr. Sherry Ferguson, along with Drs. Talpos,

Kanungo, Wang, and Liu. Again, this is probably the oldest one of the cores. It fits the mission of this division, it certainly fits within the mandate of the NCTR. The projects are characterizing not only functional behaviors but also contribute mechanistically to a great extent to our understanding. We specifically highlighted in our reports all the ongoing studies that are addressing ketamine, anesthetics, and its effects in children. I think this division has made great strides in identifying the risks that are associated with exposure of kids to ketamine, along with some of the treatments for depression and other psychiatric impairments.

The quality of the science is outstanding. We thought that the models are excellent. One of our recommendations that we had, and this applies for other divisions as well, is perhaps to increase to some extent some of the repertoire of the animal models that you have, integrate some studies in other species. One that was discussed, and you've done some work in it, actually, C. elegans.

We thought the collaborations the division has with the Mayo Clinic on anesthesia safety in kids was commendable. And we also were very favorable for continued investigation into the hepatic toxicity of acetamin and clearly, for this division, it would be important to learn

more about the neurotoxicity of acetaminophen, especially in terms of developmental neurotoxicity.

This is basically it. I think you have the report, so you can look for it. In general, when we talked about the division as a whole, we commend the division, we commend the leadership, we commend all the members. We think that the research is of high quality. It meets with the needs of the NCTR and the FDA. It has great translational value. We commend all the interactions between the different units of the DNT, outside of the DNT, within the NCTR, and certainly with other centers in the FDA.

A couple of the weaknesses, if you wish, not weaknesses, but just recommendations. As I mentioned we thought that there might be, that you might wish to consider a better way of prioritizing and reviewing the protocols that come to senior leadership for approval. We thought there's opportunities for you to increase the impact of the MRI, the CT, technology within some of the projects that are ongoing.

I think with that actually I'm not sure exactly how long I've talked, but I'll stop, and if anybody has any questions, please let me know. Again, I want to thank specifically to the individuals who were there to do the review. We were timely. I think we did a fairly good job

in reviewing the division, and I look forward to Sherry's report on some of these comments and how you are going to move it forward. Thank you very much, and I guess I'll shut up here.

DR. MENDRICK: This is Donna Mendrick. Miki, one thing we have to do is ask the SAB members to vote on whether they accept the report as written.

DR. ASCHNER: I would like to entertain a motion to approve the subcommittee report reviewing the Division of Neurotoxicology. It has to come from an SAB member?

DR. MENDRICK: Yes, the SAB members need to vote, and maybe it would be easiest if they could raise their hands.

Michael J Kawczynski: I can put a poll in here, yes or no, if you'd like.

DR. MENDRICK: Do you approve the report as written?

Michael J Kawczynski: Okay. I am going to put it out here. It will capture everyone's name with the vote, so we will know who it is.

(Voting.)

I'll keep the record for you, not to worry.

DR. MENDRICK: Miki, since you're ahead, I thought we'd go ahead and take a half an hour break now, and then Sherry will start after the break with her report.

DR. ASCHNER: Okay. I have Central time is 9:04. So let's come back at 9:30 Little Rock time, and 10:30 Eastern time.

(Break.)

Agenda Item: Response to Review

MICHAELKAWCZYNSKI: Welcome back. Just a couple little housekeeping reminders, and I love to do this, because you guys have been a great group. To all my presenters, if for any reason you do get logged out, anything happens bad, you logged out, you log back in, when you come back in, just make sure you're phone's muting, make sure you get that speaker turned off so we don't get that feedback.

On top of that, Q&A pod, we've got that Q&A pod. If you've got questions, put them in there, and then we can either call on you and stuff like that. So don't be afraid to type a question in the Q&A pod. Michael loves answering questions. So does Sherry. So do everybody else. We've got them here for two days. Let's make use of this time well.

With that, Michael, Sherry, are you both ready?

DR. ASCHNER: Okay, I just want to introduce Sherry. We'll hear now the response to the subcommittee review. Sherry, please go ahead.

DR. FERGUSON: Thanks, Michael. Thanks, everyone, for attending, too. It's Arkansas. I think you should be happy. We're having a very hot and humid August here in Arkansas. Some people call this the dog days of summer, but I can tell you, my two dogs don't enjoy it at all.

I do want to start by acknowledging the division staff in preparing the Word file to the responses as well as this PowerPoint presentation. Virtually every staff member, from support staff, from principal investigators, from our postdoctoral fellows, everyone provided input into this. So all of the good and positive things in the Word file response and this PowerPoint presentations are due entirely to the staff, and any mistakes or errors there are my responsibility.

I'm going to follow the format of previous divisions in their responses, so some of this will be a little repetitive from what Miki told you earlier, but I do want to acknowledge our subcommittee members, Miki Aschner and Greg Lanza, and our subject matter experts, Pat McConville from Invicro, who was our imaging expert and an expert in translational imaging; Ed Levin, from Duke, who was our rodent, zebrafish, and nonhuman primate expert, as well as an expert in developmental neurotoxicology; and Wei Zheng from Purdue, who was our neurodegenerative disease expert and our blood-brain barrier expert.

We had a number of goals in this subcommittee review. We really wanted to engage the maximum number of division employees. We wanted to encourage interactions. And we wanted to maximize results in projects where we wanted critique. We did this with our platform presentations, the questions after each platform presentation, our poster sessions, and then the coffee breaks where we were able to interact with the subcommittee members and others. Unfortunately, we don't have those this time.

We had three focal areas that we talked about. We had imaging, which is a relatively new technology at NCTR. Alzheimer's and Parkinson's diseases and traumatic brain injury, and these are very new focal areas in the division. And developmental neurotoxicology. This is a historical focal area since the division's inception under Dr. Bill Slikker.

We had limited time, so there were some areas of research that we didn't discuss. Most of these that we didn't discuss were protocols that were already approved or ongoing or those that were externally funded projects. I think all three of the bullet points there, though, were discussed in the poster session. That is, my study of the developmental neurotoxicology of cannabidiol, Dr. Cuevas's study of sex differences in Alzheimer's disease and

translational approaches, and Dr. Gu's brain tissue clearing studies, and I want to come back to that a bit later in this talk and give you an update as to the tissue clearing studies.

We started on Wednesday the 4th, with an overview of the division by myself, and then went directly into topic one, which was imaging. We heard presentations by Dr. Serguei Liachenko and Shusheng Zhang, and then we had a brief poster session. We had two for topic two, and four posters for topic three.

We started right back up the next morning with topic two, which was the neurodegenerative diseases and traumatic brain injury, and there we heard talks by Drs. Sumit Sarkar, Syed Imam, and Hector Rosas-Hernandez. That was followed by focal area three, developmental neurotoxicology, with presentations by Drs. Josna Kanungo, Cheng Wang, and John Talpos. Then the subcommittee adjourned.

I think the subcommittee really didn't have much of a holiday season, because we got our review in late January. It was a very quick turnaround. Some of the things that I'll quote here I know that Miki Aschner said previously, but the subcommittee said that the work carried out in the Division of Neurotoxicology is well aligned with the FDA's mission and that the division continues its

tradition of excellence in research and discovery, and our ongoing collaborations with other FDA centers are informative to the regulatory decision processes. They said the divisions reveals we have consistently increased our collaborative research across the FDA centers, and that the division has excellent scientists, equipment, and staff that produce quality research in peer-reviewed journals.

They asked about translational work and could we do any collaborative work with C. elegans or drosophila? Last fall, several members of the division, including myself, met with Dr. Tao Chen in the Division of Genetic and Molecular Toxicology, and Dr. Chen has a C. elegans lab at NCTR. It's relatively new. We've continued our discussions on potential collaborative work, and we've recently learned that Dr. Chen has ordered the WormFlow behavioral tracking system for C. elegans. And just as a brief update, it's not on the slide, but this past weekend, Dr. Chen and I have exchanged emails about a potential study that we may collaborate on looking at anesthetic exposure in C. elegans.

In November of last year, Piper Hunt from CFSAN came to NCTR and gave a seminar on her work on arsenic exposure in C. elegans. And we have continued to communicate with her because we recently completed a study of developmental exposure to arsenic in rats. So we're

comparing our results in rats to her results in the C. elegans.

Very recently we've become aware of Dr. Robert Reis's work at our local medical school, the UAMS, who works with C. elegans and drosophila in studies of the molecular genetics of longevity and neurodegeneration. If these were normal times, we would invite Dr. Reis for a visit to NCTR, a tour and perhaps a seminar, so we hope that when things get back to semi-normal, we'll be able to have him down.

The committee asked about increasing the understanding of the mechanisms by which a potential drug works to treat brain diseases, and they specifically mentioned the monoclonal antibody aducanumab. This is a biologic that was recently submitted to the FDA with a biologics license application, and the FDA has given it priority review. It does seem to be well-tolerated in Alzheimer's patients.

Dr. Sarkar has a colony of a double transgenic rat model of Alzheimer's disease, and that would be ideal to decipher the mechanisms by which aducanumab interacts with and crosses the blood-brain barrier to potentially remove plaques in the brain. Of course, in subsequent protocols, several potential Alzheimer's drugs with the

ability to penetrate the blood-brain barrier will be included for study.

This is something that Miki said earlier this morning about external review of our protocols, and I'm not sure if the subcommittee members were volunteering to review our submitted protocols or not, but I've been at NCTR now for 30 years, and at one time all of our submitted protocols were externally reviewed. The principal investigator of the protocol would submit names of people from academia, industry, or other government agencies as reviewers, but we currently use subject matter experts within the agency for review now.

The first focal area was our imaging work, and the subcommittee said the potential impact of our imaging research and services within NCTR is significant and unambiguous and that the use of our T-2 relaxation time as a biomarker of neurotoxicity has progressed. And in fact, Dr. Serguei Liachenko has almost achieved full approval for his protocol, which will expand the knowledge of the mechanisms that are associated with drug-induced neurotoxicity and possible pathways to prevention. More specifically, his study will describe the developmental changes in T-2 as well as validating the biomarker performance with higher resolution images.

The committee commented on our PET imaging, noting that the quality of the PET imaging data is high. And they mentioned the blood-brain barrier penetration of the compounds. And we've been aware of this potential confound for a while. To give you an example, much of Dr. Zhang's work has been with the use of pediatric anesthetic. If a postnatal day 7 rat is exposed to sevoflurane, for example, then it is typically imaged one week, two weeks, three weeks later, in the PET imaging instrument. She uses radiotracers. Many of those radiotracers are apoptotic, they bind to apoptosis, and in her studies, she's seen increased concentrations and increased retention of those radiotracers that are targeted for apoptosis in the animals that were previously exposed to anesthetics. So the question is, did the anesthetic truly cause increased apoptosis in the brains of those animals, or did the anesthetic actually alter the blood-brain barrier of those animals, making it perhaps more permeable, such that those animals showed increased levels of the radiotracers?

To better understand the potential of developmental exposure of these anesthetics to disrupt or alter the blood-brain barrier, Dr. Zhang is working on an addendum to examine this, and I want to give you a little bit of detail about the addendum, because it is still in progress. Not the work, but just the submission of the

addendum. She's going to use sevoflurane exposure on postnatal day 7 rats, and that will be identical to her previous study, and then this particular addendum will be done in a staged manner.

Initially, the expression of those tight junction proteins such as claudin 5, occludin, and ZO-1, will be measured at the same ages as she previously did her PET imaging. Those ages would be postnatal day 14, 21, 28, and 35. If that result shows differences in the expression of those tight-junction proteins, then the blood-brain barrier permeability will be measured with FITC-dextran extravasation at the same ages. To give you an idea of what this might look like, these are images from a 2017 article. On the left you see an intact blood-brain barrier of a control rat, and on the right, you see the blood-brain barrier of a rat that was treated with methamphetamine showing a much more permeable blood-brain barrier.

The committee asked about enhancing our imaging analysis capabilities, and we have a number of imaging modalities at NCTR, including MRI, PET/CT, electron microscopy, MALDI, and other forms of microscopy. So all of our imaging modalities are discussing the formation of a joint image analysis center that would implement the most current algorithms, paradigms, software, and expertise in order to optimize and streamline our image analysis. We've

also been in conversations with Dr. Angel Paredes, who runs our electron microscopy here, and our division has volunteered to pay for half of a particular software that will be used for our brain assays. I think the software is called Dragonfly. I'm not very familiar with how this works with the electron microscope, but I've seen images that it can do and it looks very, very good.

The committee asked about artificial intelligence modeling and prediction and machine learning. We have started discussions with the Division of Bioinformatics and Biostatistics in order to develop our AI approaches to increase image quality and automate our diagnostics.

The committee asked about using longer-lived PET isotopes and that this might be appropriate for in vivo tracking and biodistribution imaging, and historically we have not used these. We have learned, however, that our radioligand manufacturer, 3D Imaging, can make these for us. So these longer-lived PET isotopes would expand our imaging applications, especially where there's a molecule that can't be easily labeled with F-18.

Those radioligands would cause increased radiation dose, and there would be some issues related to waste handling. And then with all of our PET images, the animal is typically returned to the colony, returned to its home room and its home cage, so there may be some issues

with adequate shielding, but we could probably deal with those. So this is possible in our future.

The committee noted that we have limited technical staff and recommended that our personnel resources be examined. In March of this year we hired a very, very strong experienced MRI technician as a support staff scientist. His family was in Maryland. He came to us from CDER. And then COVID-19 happened, and he took a reviewer position with CDRH in order to be closer to his family. So that particular position remains open.

As you might imagine, hiring at this time is incredibly difficult. We are prohibited from traveling anyone in for a physical interview. Even for those that might come to the NCTR or take a job offer, accept a job offer, traveling and moving is very uncomfortable for many, so that open position is still open. The search continues.

The committee asked about sharing of FTEs between labs. Typically our imaging labs are feast or famine. That is, we have many animals running through the MRI within a couple of weeks, or many animals running through the PET within a couple of weeks. We have hired a new postdoctoral fellow to assist with the microPET/CT as needed. We have two support scientists currently dedicated to MRI work and that one open position. So we try to

adjust the flow of personnel depending on what the work needs.

The second focal area was our Alzheimer's disease, Parkinson's disease and traumatic brain injury work. The committee noted that we were focused and off to a commendable start, that we had significant accomplishments in a relatively short time period, and that the topics studied are timely and meritorious. They noted that the process of neurodegeneration and its treatment is key to the mission of the FDA.

The committee noted that we should be investigating the relationship between behavioral function, mechanistic imaging, and molecular mechanisms. I did have, four years ago, an addendum to Dr. Sarkar's protocol with his double transgenic rat model of Alzheimer's disease, and what I really wanted to look at there was preweaning behavioral development, early landmarks, eye-opening, incisor eruption, et cetera, and prepubertal play behavior.

Well, in the process of that addendum getting approved, the colony was moved at least two to three times to different buildings. Some of those buildings had construction noise, and it significantly affected the breeding of those animals. We purchased additional breeding pairs to bring those numbers back up, and we hope

to continue that work looking at the early behaviors of that transgenic rat.

His current protocol -- and actually this is a little bit -- he has already assessed the cognitive deficits in the cerebral amyloid angiopathy mouse. And while there were mild cognitive impairments in that CAA mouse, the most striking differences were the sex differences in there. And these haven't been described in that mouse model before, which really just highlights the idea that we always should look at both sexes. In his double transgenic Alzheimer's rat model, he has a new postdoc who is going to look at behavioral progression of the disease.

With regard to mechanistic imaging, we have used tau and amyloid beta-based microPET methodologies and MRI imaging, and will continue to include those in future protocols. Those protocols often include behavioral assessments as well. One of the things I learned in preparing for this talk that I didn't know is that in the past, Dr. Zhang has used F-18 florbetapir, or Amyvid, and this compound binds with high affinity to beta amyloid plaques. It easily penetrates the brain, and she used it in Alzheimer's disease transgenic mice. Those that were eight months old, the transgenic mice, had increased uptake and retention in the frontal cortex, but that wasn't true

at three months of age. So this really gives us a starting point for future work.

With regard to molecular mechanisms, this was an oversight on my part in the December SAB. I neglected to mention that several of our protocols include molecular mechanistic endpoints. Several of our ongoing protocols are investigating gene expression, the microbiome, and microRNAs. We've also recently purchased the Indica Lab's Halo microglial activation module, and this is the gold standard image analysis platform to detect activated and inactivated microglial cells and non-microglial cells. This software will do cell classification and counting, and it precisely measures cell processes and branchpoints per cell. This will greatly speed our data analysis.

They recommended that our researchers adapt dynamic microfluidic models and human induced-pluripotent stem cells in our proposed neurovascular projects. We have an ongoing protocol in the division with the PI of Dr. Sayed Ali, and that will use a commercially available microphysiological system known as Synvivo, and primary human brain endothelial cells and neurons to model the blood-brain barrier. This will allow for a better model of the in vivo morphological and physiological characteristics of the blood-brain barrier in an in vitro setting.

Initially Dr. Ali will test the effects of ketamine and propofol.

Continuing with that line, we have a recently approved protocol with a PI of Dr. Rosas-Hernandez to construct a brain-on-a-chip model, and this will be a model of Alzheimer's disease, and it will use neurons, astrocytes, and brain-like endothelial cells derived from human induced-pluripotent stem cells from several different populations, including cognitively healthy subjects, that would be the controls, and those with familial mutations of Alzheimer's disease, including APP, PSN1, and PSN2, as well as those with the APOE4 allele. Successful implementation of this model will increase the FDA's expertise to better understand and interpret data that are submitted by industry on the safety and efficacy of drugs, biologics, or medical devices using human induced-pluripotent stem cells and microfluidic models. It will also allow the FDA to conduct studies about potential safety concerns of FDA approved drugs, biologics, or medical devices for the treatment of Alzheimer's.

The committee noted that it wasn't clear how the TBI work fits into the needs of the FDA, and again, this was a total oversight on my part. This was certainly not due to Dr. Rosas-Hernandez's presentation. So I just want to spend a minute to kind of clear that up. You're aware

that two years ago the FDA approved the first diagnostic test to evaluate concussions or mild TBI without the need for a CT scan. And of course future diagnostics will also require FDA approval.

So if we have a TBI model that is explicitly designed to induce mild, moderate, and severe TBI, this will allow the agency to have a reliable and reproducible means to study, validate and qualify not only diagnostic but also prognostic and predictive biomarkers for specific injury severities. Those biomarkers are likely to be fluidic, we all know that. But they could also be medical devices or improved imaging methods. As just one example, CDRH is looking at a portable EEG device that can go into war zones to diagnose a TBI. Our EEG findings in a validated TBI rat model could establish potential biomarkers for brain injury monitoring.

Then, of course, we're all aware that FDA-approved and nonapproved drugs are taken by those who will get a TBI in the future, those who have a history of TBI, those who might be being treated at the time for a TBI. So let me just give you an example of how we envision this work progressing. You could have a population of adults with attention deficit hyperactivity disorder, who are taking methylphenidate and then another population that is not taking methylphenidate. What are the differences in

those two populations if they get a TBI? Should the TBI be treated differently? Are there different biomarkers for the TBI? So it's really not clear what differences in those populations might be. We don't know what the potential toxicities in those different populations might be. So successful implementation of this model is going to benefit all of the centers. They can then request studies on the effects of specific drugs, biologics, medical devices, both before and after a TBI.

Then of course, I think that Dr. Rosas-Hernandez also talked about the future of this particular model in terms of neurodegenerative diseases resulting from one TBI, consecutive TBIs, are there sex differences, and so on.

The committee asked about our research on exosomes, recommending that we include the analysis of bioactive molecules other than amyloid beta, such as alpha synuclein. And Dr. Rosas-Hernandez has a protocol that will examine postmortem human brain tissues, serum, and CSF for the role of exosomes in Alzheimer's progression. Those exosomes will be isolated from several brain regions, and the cargo analyzed to determine oligomeric forms of amyloid beta, and oligomeric and phosphorylated forms of tau.

He also has another protocol to evaluate the release of alpha-synuclein in exosomes after in vitro simulated TBI, in control human dopaminergic neurons, and

those that have been treated with MPP-plus to simulate Parkinson's disease.

The committee noted that we were stretched too thin with many directions. We do have, the scientists in this particular focal area, do have a lot of areas of interest, but those aren't necessarily reflected in the hours or the funding that we put into particular studies. Our core focus will always be those that directly impact regulatory policy.

I do want to note that the division has now four open positions related to the three focal areas. The first position is a principal investigator level position for a neuropathologist. We have offered that position to someone who has accepted, and we hope to have that person on board in November or December. To assist that PI, we also have an open position for a support scientist to provide additional support for the neuropathology work. And then of course, we have our support scientist position to provide MRI support. And we just learned last week, as well, that our administrative assistant will be leaving next week, so we have that position open.

So I'm going to make a shameless bid here to the audience, that if you know of anyone that might fit any of these positions to please get in contact with me. Again,

hiring just remains incredibly difficult during these times.

They asked about studying neurobehavioral processes that are impacted by chemical exposure. As the head of the neurobehavioral lab here, I can tell you that our labs are in high demand, both within and across divisions, and in part this is because we adhere to EPA and OECD guidelines, which specify a minimum of 20 per sex per treatment group. We avoid litter compounds, so siblings are not counted as separate subjects. And for this reason our large studies often take all of our available capabilities for 12 months or more. For example, the developmental exposure to CBD study, which is ongoing, and our recently completed developmental exposure to arsenic. But typically, when we have smaller acute exposure studies, they can be worked in alongside these large studies.

We have recently completed funding 16 additional rodent operant chambers, which will expand our abilities to assess various cognitive functions, and this will also allow for easier assessments for those smaller acute exposure studies, but still allow those studies that require long-term assessments to continue.

Focal area number three was our developmental neurotoxicology work. The committee noted that research in this area hits the core of the interests of the FDA and

that seminal research by investigators in this core on the developmental neurotoxicity of anesthetics is laudable, and our collaboration with the Mayo Clinic is commendable. One particular collaboration that I neglected to mention in the December SAB was our collaboration with the Icahn School of Medicine and the National Institute of Perinatology in Mexico City. That collaboration has recently produced a manuscript that will be submitted soon on prenatal exposures to metal mixtures and later reward motivation in children.

Our introduction to the developmental neurotoxicology work didn't mention personnel arrangement or facility needs. Again, an oversight on my part. Adequate personnel and staff are often in short supply. So when an FTE or government position becomes open, and this can be due to retirement or leaving to another agency, our leadership evaluates the research areas and attempts to fairly distribute personnel.

I want to say something about facility needs. Our annual maintenance contracts for the two MRIs are $74,000. The microPET/CT instrument is approximately $125,000. Maintenance of those two instrument consumes 18 percent of our annual total division budget. One PET radiotracer synthesis can cost anywhere between $3,500 and $15,000. One synthesis can typically image three rats. So

our imaging sucks up a large portion of our budget, and I think that one of the things that the committee recommended is that we try to increase our use of our imaging facilities, and we definitely need to do that, since they cost so much.

They asked about using minipigs to complement our nonhuman primate research, and as Miki knows, we actually have a history of working with the Yucatans. Our previous studies have described environmental enrichment preferences and performance of operant tests. We've had some discussion about potentially bringing the species back to NCTR or starting collaborations with those using this species at our local medical school. One of the things I do want to note is that young adult minipigs can be imaged in our 4.7-tesla MRI.

Dr. Talpos has initiated a collaboration with Paul Morton at Virginia Tech. Dr. Morton is going to look at physiological endpoints in minipigs, but he would like to collect behavioral data on that species as well, and Dr. Talpos is our behavioral expert, and he will provide recommendations on assessments.

The committee said that we should establish a close collaboration with the Division of Bioinformatics and Biostatistics. When possible, we do such collaboration, and as one example, Dr. Fang Liu has collaborated with Dr.

Binsheng Gong in the Division of Bioinformatics and Biostatistics, and a coauthored manuscript is now under review. She's also developing a collaboration with DBB for work in which the brains of infant monkeys that were exposed to desflurane, an anesthetic, were examined for microRNA and messenger RNA. The researchers in DBB will analyze the data and correlate the regulatory roles of the microRNAs with messenger RNA expression.

The committee noted that nonhuman primates are useful to evaluate psychiatric drugs, and indeed they would be. Our approach, typically, with any potential neurotoxicant is to initially conduct such studies in rodents, followed by the studies in nonhuman primates when necessary. As one example, you're probably aware that the FDA recently approved the use of esketamine as an antidepressant, but it's not approved for use in anyone under the age of 18. It's likely that that sort of use may be necessary.

So to study the effects of that potential neurotoxicity, Dr. Talpos has a protocol to look at the effects of early and adolescent acute ketamine exposure, in rats, to determine the potential neurotoxicity of ketamine use as an antidepressant in children and adolescents. If there are indications in that rodent study, they can be replicated or extended in nonhuman primates. But certainly

given the increased use of antipsychotics in children this could be a very promising area of research.

The committee said that with each project it's recommended that there be a dual focus on tests of potential neurotoxicity, as well as investigation of the mechanisms of neurotoxicity. One of the things that's not in here as a bullet point that I did want to mention, is that this is precisely how Dr. Josna Kanungo's studies are conducted. Zebrafish embryos are exposed to a potential neurotoxicant, she sees a phenotype, and then she goes after the mechanisms.

And several of our recent protocols have actually been staged in this manner. I had a protocol a couple of years ago to look at the behavioral alterations that were produced by a single episode of sevoflurane anesthesia in adult rats. There were no behavioral deficits due to sevoflurane, but had there been, we had several additional phases to that protocol to investigate the mechanism. So for the most part, the neurotoxicity potential of a compound is first determined, followed by a study of the mechanisms.

I want to spend just a minute, and I'm glad that Dr. Aschner brought this up as a question to Bill, spend a minute on the impact of COVID-19 on our division. In March of this year, all FDA employees were instructed to

aggressively maximize telework, and this includes our postdoctoral fellows. This directive of maximizing telework is still in place. Some of our division employees are in high-risk categories and to this date continue to telework 100 percent.

So for some studies lab work has been minimal, and there has been very intense planning regarding the startups and continuation of studies, and that intense planning involves how many employees need to be on site, how many employees need to be together in a lab, together in the animal areas, together in offices?

Our division is very fortunate that with one exception all of our staff have an individual private office, including postdoctoral fellows. But I certainly don't want to make it sound as if we haven't made any progress at all. We hired four new postdoctoral fellows since the December subcommittee review. All of those people had physical in-person interviews prior to COVID-19.

I do need to point out that there is a typo in the second bullet point. Since the December SAB, there have been nine -- not 17 -- there have been nine manuscripts submitted for review and an additional 10, not nine, publications. So we have a total of 19 manuscripts that are either under review or have been published since our December SAB meeting. Now we're only eight months into

the year, but to give you an idea, we're currently at 19. Last year our total was 11. The year before that, 2018, our total was 22. So we're well on-track to surpass, and perhaps set a record, for number of manuscripts this year.

Then I mentioned earlier that I wanted to update you on Dr. Gu's progress on the tissue clearing. Now it's probably where I need that pointer. Shown here are diagrams of sagittal sections of the brain. Each diagram represents a different neurotoxicant. Where the pointer is right now, that shows amphetamine. The brain regions that are shaded in red show the brain regions that would be affected by amphetamine. Right below that, MK-801, 3-NPA, and PCP.

The blue horizontal lines are the classical brain slices that are taken to look for neuropathology. In the instances of amphetamine, MK-801, 3-NPA and PCP, the three traditional brain slices would certainly detect neuropathology. But now move over to the far right. This first diagram shows the effects of MPTP, alcohol, carbonyl sulfide. So if you take your three traditional brain slices, you would not see neuropathological effects due to MPTP. But we know that MPTP has tremendous functional effects, and the brain regions that are affected by a neurotoxicant may be very small, but the minor damage could have a very significant functional impact. And we know

that's true with MPTP, and the brain regions that might be vulnerable to a potential neurotoxicant are sometimes unpredictable.

So in 2011, the National Toxicology Program adopted a new protocol moving from three slices to seven. And I think we'd all agree that seven is better than three. But what about 60? Is 60 better than seven? Sure. Any of those small regions that were affected by MPTP, if you took 60 sections, you'd see the effects. But of course, doing something like this is very time intensive. There are some core programs that will put the images together for you, like Neurolucida, but the time involved in slicing the brain, in staining the brain, in imaging those slices, is very, very tremendous. So this is where tissue clearing comes in.

Tissue clearing makes possible investigating large intact tissue volumes, such as an intact mouse brain, and you can do it with cellular resolution. The clear tissue retains the capacity for immunolabeling as well. So I want to give you an update on Dr. Gu's progress here. He's looking at the three typical methods for tissue clearing. Shown in the top left is a mouse brain with no tissue clearing. Shown in image B is an aqueous hyperhydrating based method, that's also known as the cubic method. Shown in image C is an organic solvent-based

method. This is known as the uDISCO method. And image D shows a hydrogel embedding-based method, which uses a hydrogel monomer to crosslink macromolecules first, and then the lipids are removed.

And that's really the whole focus of tissue clearing is to remove the lipids, to make the tissue almost glasslike-clear. It leaves behind -- of course he's still working with these, these are just preliminary images -- it leaves behind a transparent glasslike brain. The vasculature, the cells, and even the synapses are all intact. This technique has been used to look at 3D visualization of amyloid plaques and tau in Alzheimer's disease mouse models.

He's moving forward with this. Once those three particular methods are perfected, then he will move to treating the animal with a neurotoxicant, and the first one will be kainic acid. Everyone knows kainic acid is a neurotoxicant, and we all know what brain regions it affects, so the mouse will be treated with kainic acid, and then tissue clearing will happen, and then he will move to a fluorescent tracer, something like Fluoro-Jade.

I think that this is a really novel method, and Dr. Gu is one of our best methodological scientists in the division, so this could impact drug development by allowing easy visualization of whether a drug can cross the blood-

brain barrier, and I'll use a really terrible pun here, but in my opinion, tissue clearing is clearly a better way to evaluate neurotoxicity.

I mentioned in our December SAB that we had started MRI images of adult nonhuman primates. Those nonhuman primates were involved in a study of methylphenidate, where we had a control group, a low-dose group, and a high-dose group. They began on methylphenidate when they were two years of age. They were on methylphenidate for 12 years. And then they had a two-year washout period. After that two-year washout period, we MRI imaged them.

Shown here are average T2 maps for each group, and although there were no -- Dr. Liachenko looked at 22 different brain regions here, and although there were no differences with methylphenidate, the image quality and the co-registrations were very good.

One of the most exciting things I think that the committee mentioned is suggesting that we establish an interdisciplinary group with a blood-brain barrier focus. We're very excited about that. Blood-brain barrier research remains a strong focal research area in the division. We have a postdoctoral fellow that I think many of you met at December's meeting, Dr. Andrew Shen, and we have converted him to a staff fellow. He comes to us with

extensive experience in blood-brain barrier assay methods. Some of our current projects in the division that are looking at blood-brain barrier are Dr. Cuevas's project on the assessment of sex differences in beta-amyloid binding to blood-brain barrier transporters in a transgenic mouse model of Alzheimer's, Dr. Sarkar's assessment of vascular function in a transgenic rat model of Alzheimer's, and Dr. Rosas-Hernandez's protocol with a microphysiological system, or that brain-on-a-chip, using patient-derived human induced-pluripotent stem cells, to model blood-brain barrier dysfunction, and this is a collaboration with Emulate.

I believe that was my last slide. The Word file that we gave to the subcommittee contains much more detail than what I presented here in this PowerPoint presentation, but I do want to end by acknowledging the subcommittee members. They have provided incredible input, we were very gratified to know that our research is well recognized, well acknowledged, and the critiques and the reviews that they gave us, we're going to put to use. So I'll stop there and answer questions.

DR. ASCHNER: Thank you, Sherry. I think this was a very thorough actually response. Thank you for sharing some of the new studies that you're doing. I personally don't have any specific questions. I would just say that

these are recommendations, and just keeping in mind one thing that we said, you don't want to stretch yourself too thin. Hopefully, and you did, you should read our recommendations, but it doesn't necessarily mean that you have to implement them all in the first year. This is just sort of a roadway for you to think about how you might want to incorporate new technology and address some issues, some concerns, and so forth.

So I'm happy with the response. I know Greg is online, he was on the committee, the other members are not, obviously, because they were just for the subcommittee meeting, they're not on the -- I can hear somebody in the background typing.

MICHAELKAWCZYNSKI: We have Steven Stice, you have your hand up.

DR. STICE: Yes. Really nice work, Sherry, and your group does a fantastic job in giving us an update on all the things that you're doing. I guess I would just echo what Miki is saying about spreading yourself too thin. I assume that you've thought about a model where you guys are the experts, you know a lot about these model systems, what the physiology and toxicology around systems. There are contract research organizations out there that can fulfill certain types of assays, types of things, but you are the brain trust. Have you ever thought about being

sort of that connector between CROs, where you can provide the expertise that can guide study designs that may be some outside groups can do?

And then, I really like the idea of using your imaging capabilities to the utmost. Maybe that will allow you to concentrate on, with the using the tools that you have in hand on very specific projects. Just a thought.

DR. FERGUSON: Right. I would first have to find out is it ethical or legal for us to talk to CROs to offer advice or expertise? As a federal agency, I'm always a little bit unclear as to who we can talk to. It's a good thought, though. It's a very good thought.

DR. STICE: I am not sure. I would not recommend giving advice to the CRO, but if an agency comes to you with a problem, you design the experiments around that problem, and then if there are people that can do specific things helps your group, it might just increase your throughput, and you can concentrate on the more complex types of activities. Just a thought.

DR. FERGUSON: Right. Now I understand, yes.

DR. ASCHNER: Thank you, Steve and Sherry. Are there any additional questions for Sherry?

Okay, hearing none, thank you again for all the work that the division is doing. As noted, we were all impressed. There's always room for improvement, that's why

we were there, and we look forward to reviewing your division in five or six years again. And good luck.

DR. FERGUSON: Thank you, Miki.

DR. ASCHNER: Thank you.

I guess we are going to go through the different centers now, so I would ask each of the speakers, I'm going to introduce you, but if you can just mention just briefly your responsibilities within the division or within FDA in general, we'd appreciate it. Each of you, I believe, has about 20 minutes, half an hour. We're ahead of schedule. I won't stop anybody. If you need an extra five minutes, it's not a problem today.

The first speaker is Dr. Denise Hinton, and she'll provide us with a statement from the Chief Scientist.

Thank you, Denise.

Agenda Item: Statement from the Chief Scientist

RADM HINTON: Thank you. Good morning, everyone. I am Rear Admiral Denise Hinton. As always, I am very pleased to be able to join you this morning, to hear more in detail about the work NCTR and all of you are doing. And I must say, I'm always impressed by the scope of research underway at NCTR in support of FDA's work and by NCTR's leadership.

Today, as COVID-19 rages through every aspect of our lives, your work is more critical than ever. I want to talk for a moment to express our deep appreciation for all the work being done by NCTR laboratory researchers, and by the lab workers across the agency. All of you, like other frontline workers, are facing particular challenges in your efforts to support the development of COVID-19 targeted diagnostics, therapeutics, and vaccines. I also want to thank our Office of Laboratory Safety, and the staff there, who have been working hard to ensure the safety of our labs and the surrounding communities, as well as their collaboration with all the centers and offices to develop internal laboratory safety guidelines for personnel working with the virus.

After listening to NCTR's achievements over the past year, I think you have to agree with me, NCTR is making remarkable contributions both within the agency and with our domestic and international stakeholders, and I want to take a little time to note those efforts. I sincerely regret that time won't allow me to highlight NCTR's significant role in this COVID-19 response as much as I'd like to, but I'll just highlight a couple.

NCTR was one of the first entities in FDA to initiate research into COVID-19 back in March. Since then, NCTR has been deeply engaged in activities supporting FDA's

regulatory role, conceptualizing 26 research proposals to combat the COVID-19 pandemic, of which five studies are ongoing or in development. One of NCTR's first projects was development of an approach that can be used to rapidly indicate effectiveness of COVID-19 therapeutic treatment. This project is expected to continue into FY2021.

NCTR has also recently initiated an artificial intelligence project supporting efforts to effectively treat COVID-19 patients. This project is supported by OCS's, Office of the Chief Scientist's, Medical Countermeasures Initiative, and aims to systematically survey and prioritize approved or investigational drugs for their potential use to treat COVID-19.

Another ongoing study is testing for the presence of SARS coronavirus-2 in wastewater in the central Arkansas area. Early detection and continuous routine monitoring of the virus in the community can help federal agencies implement methods to limit the spread of the virus, decreasing the burden on healthcare facilities, which is an immediate impact on public health. It's a public health benefit.

Unfortunately, the coronavirus pandemic is disrupting public health services across the country, threatening to undermine what progress we've made in the opioid crisis. But we must not relent in our efforts to

address that crisis. NCTR continues doing its part with several ongoing projects related to opioid addiction and toxicity potential that will help inform FDA's opioid action plan to provide comprehensive guidance for reestablishing safety standards for these products.

One NCTR-CDER collaboration is using computational models to assess the structure of addictive chemicals. This project should give us a better understanding of the structural requirements associated with a strong addiction potential and may prove useful in prioritizing the testing of chemicals with strong addiction potential like synthetic opioids and cannabinoids, thus shortening the FDA regulatory review process.

In another NCTR-CDER collaboration, scientists are conducting research to assess perinatal opioid exposure, a concern FDA shared in its perinatal-related drug safety communications.

This brings me to a subject very close to my heart, and that's NCTR's Perinatal Health Center of Excellence. As Bill mentioned, the Perinatal Center of Excellence held its first annual workshop at the White Oak facility to review research progress, funding three new research studies, and bringing the total of ongoing projects to 17. The unique public health challenges facing this understudied population have been on full display

during the COVID-19 pandemic, and as the disease's effect on pregnant women in particular have been often fraught with complications and sometimes death. The data on pregnancy and COVID-19 are far from complete, but to date they suggest that although the virus is unlikely to affect early fetal development, pregnancy can make women more vulnerable to the effects of severe COVID-19. This is due in part to the way the immune system calibrates itself during a pregnancy, and because the coronavirus attacks the lungs and the cardiovascular system, both of which are incredibly stressed during pregnancy.

As many of you have heard so many times, you've heard me say it and many others, my office has been, and will continue to be, fully committed to raising awareness around the scientific research being conducted at NCTR, and about its impact on our regulatory decision-making, and supporting NCTR in its work to protect public health and advance the innovative tools and approaches that are critical to FDA's ability to predict risk and efficacy.

We're doing this in a number of ways. NCTR's research continues to be a regular feature of the OCS-sponsored FDA Grand Round webcast. This was launched in 2016, the goal of which has been to raise the visibility of FDA's research and its effects on laboratory activities. On Thursday, August 13, NCTR's Dr. Anil Patri gave a Grand

Rounds presentation on the OCS-supported Nanotechnology Task Force report, which was published in July and is now available on FDA's website for viewing. Dr. Patri highlighted FDA's advancements made over more than a decade, including nano research, the issuance of guidance documents, development of standards, formation of domestic and international collaboration, and emerging challenges in regulatory science. My office has also supported cross-agency nano research since 2010 through the management of the collaborative nanotechnology intramural grants program, we call CORES.

Because regulatory science research in nanotechnology is inherently multidisciplinary, the CORES grant program is designed to promote cross-center and external collaborative research opportunities to address high-priority FDA regulatory science nanotechnology research needs. Results of research from CORES grants have provided data to support the publication of guidance documents in the development of regulatory science tools like assays, assessment methodologies, and test protocols that FDA uses to evaluate nanotechnology in FDA-regulated products. Moreover, the CORES program has increased collaboration across FDA and continues to strengthen our relationships with academia, industry, and other federal agencies.

Although it seems like a lifetime ago, exploring potential avenues for collaboration in greater alignment with scientific priorities was one of the key goals of the first ever NCTR science day held at White Oak on March 6. The OCS-sponsored event also gave center directors and scientists here at headquarters the chance to see in person the range and scope of NCTR's cutting edge research and meet with NCTR's leadership and research staff, and we expect this to be the first of many annual NCTR Science Days, either in person or virtual.

As many of you know, NCTR leadership can be found just about everywhere in OCS-led cross-agency efforts, and their contributions are often central to some of FDA's highest scientific priorities. NCTR's Dr. Bill Mattes chairs the biomarker working group. Dr. Donna Mendrick serves in a variety of areas, including as acting chair of FDA's Center Science Council, cochair of the alternative methods working group, and chair of the artificial intelligence working group.

The artificial intelligence working group has invited expert speakers from within and outside the agency and has organized an educational subcommittee that is working with Harvard-MIT's Center for Regulatory Science to offer AI educational series. On the alternatives spot, NCTR is expanding efforts using microphysiological systems,

as you just heard Dr. Ferguson mention, and is positioned to test side-by-side this new technology alongside gold-standard proven studies.

The alternative methods working group is also sponsoring an FDA webinar series for agency scientists, inviting developers to present their cutting-edge technologies for disease modeling, efficacy and safety. There's more important information about this available on FDA's website.

The alternative methods working group will be publishing its report to Congress on agencywide efforts in this area later this year, and we can't predict next May's public FDA science forum, which will also be supported by OCS and include all of you. NCTR has been engaged with all FDA centers and offices in the forum's organization, and in shaping its key topic areas, which will include tools to predict toxicity and the efficacy of FDA-regulated products in humans and in animals.

Finally, I'd like to recognize the important role NCTR plays in promoting global harmonization and the standardization of regulatory science in its work with our international partners. Under Dr. Slikker's leadership, NCTR established the Global Summit for Regulatory Science in 2011 and is bringing together leaders from nine countries and the European Union each year to focus on

regulatory science research. These partnerships, like the Global Coalition for Regulatory Science Research are focused on modernizing safety assessment through global exchange, training, and collaborative research with toxicologists and other scientists worldwide. This year's global summit, originally scheduled to be held in Bethesda from September 20 to 30, taking place virtually due to COVID-19, of course. So for the first time ever we all will have an opportunity to listen to the 60-plus speakers from FDA, and around the globe, on emerging technologies for regulatory applications. The recorded sessions will be available as of September 23, and then on September 30 there will be a live Q&A session, just to reiterate.

In closing, I want to thank all of you for your commitment to public health and for your work you do in support of FDA. Thank you also for the extra effort that went into pulling this outstanding program together virtually. I hope all of you will stay safe and well.

Now I'll turn the podium back over to the centers, and I look forward to hearing the centers' perspective about the other toxicology projects under way.

Thank you.

DR. ASCHNER: Thank you, Dr. Hinton. We appreciate your comments. Are there any questions for Dr. Hinton?

(No response.)

Okay, hearing none, again, thank you for the introduction and for describing the mission. We appreciate it, and we'll move on to our next speaker, which is Dr. Braunstein. You see the title, and she comes to us from CBER.

Go ahead, Emily.

Agenda Item: FDA Center Perspectives

DR. BRAUNSTEIN: Thank you for having me talk to you today. My name is Emily Braunstein. I'm the CBER scientific program manager. I work with Dr. Carolyn Wilson who is the associate director of research at CBER. I'm going to talk to you today on the research that we have going on in the Center for Biologics Evaluation and Research, and how we work together with NCTR.

Within Center for Biologics, we have four offices that conduct research. Three of them are our product offices. OVRR, the Office of Vaccines Research and Review, OBRR, the Office of Blood Research and Review, and OTAT, the Office of Tissues and Advanced Therapies. The fourth office that does research is the Office of Biostatistics and Epidemiology. They provide statistical, epidemiological, mathematical modeling support for the product offices, as well as also doing post-market surveillance of CBER regulated products.

CBER regulates a number of very diverse biological products. For example, in OVRR, they regulate over 1,400 allergenic products, vaccines, both preventative and therapeutic, and also live biotherapeutic products. In OBRR they regulate blood and blood products, HIV diagnostics, and devices related to biologics. In OTAT they regulate plasma-derived proteins and recombinant derivatives, cell and gene therapies, regenerative medicine therapies and xenotransplantation products. You can see that we regulate a very diverse number of products.

CBER advances the scientific basis for the regulation of our biologics, ensuring our projects are strategically aligned for four research goals. We evaluate these goals every two to three years to ensure that they're aligned with center, agency, and departmental objectives. I'm not going to read through all four of the research goals, but as you can imagine, all of the collaborative work with NCTR really falls under goal two, which is developing and assessing nonclinical models and methods predictive of clinical performance with respect to toxicity and effectiveness.

To go along with the diverse nature of the products regulated by CBER, our scientists also need to have a wide variety of expertise. So we have researchers proficient in many applied technologies such as NMR, mass

spec, flow cytometry, high-throughput sequencing, but our scientists are also highly skilled in microbiology, immunology, biochem, cell biology, microphysiological systems. And we also have talented in-depth scientists working in bioinformatics and biostatistics.

The CBER research program has a state-of-the-art research facility at White Oak. We have a few core facilities. We have flow cytometry, which allows researchers to do cell sorting and analysis. We have this under BSL-2 and BSL-3 conditions. We also have confocal and electron microscopy core facilities. We have a biotechnology core facility, which has all these technologies listed, and we also, CBER also provides bioinformatics support through HIVE, which stands for high-performance integrated virtual environment. It's a cloud-based environment that's optimized for storage and analysis of extra-large data like next-generation sequencing data.

We also have a state-of-the-art vivarium that includes an imaging facility with an MRI, digital x-ray, IVIS, ultrasound, microCT. We have ABSL-2 and ABSL-3 labs and procedure rooms, as well as a transgenic derivation facility.

CBER advances the regulatory science by collaborating with scientists from all over the country, all over the world, and from all different sectors,

including government agencies, industry, academia, and others like nonprofit organizations. We currently have 37 collaborations with scientists within the agency. Fourteen of these are collaborations with NCTR. There's almost an equal split where CBER is leveraging NCTR to support our research projects and vice versa.

I would like to give you a brief update on some of the ongoing collaborations we have between CBER and NCTR. Although these slides talk about the progress since last year, some of the progress on these projects may have slowed down due to the COVID pandemic.

An analysis, when we were looking at the current research projects that we had between CBER and NCTR, we found that they mainly fell into three different categories. Metabolomic and lipidomic analyses, evaluation of alternative in vitro systems, and genomic evaluations.

The first project is a collaboration between Dr. Carlson from CBER and Dr. Sun and Dr. Berger from NCTR on metal metabolomic and metagenomic analysis of fecal samples. I think the main intent of the project is identify biomarkers of protection to C. diff infections that could be found in the microbiota. Dr. Carlson began developing an animal model to look at the potential markers of efficacy for FMT, fecal microbiota transplantation, with regard to treating C. diff infection. He found that MAIT

knockout, which is mucosal associated invariant T-cell knockout mice, are resistant to C. diff infection and that the microbiota from these mice confer resistance to wildtype mice.

So in addition to metagenomic studies they did, they're also working to identify metabolic byproducts from the microbiota that could be associated with this phenotype. The collaboration with NCTR is really important because we're able to leverage the unique capabilities that NCTR has to develop assays to evaluate metabolites, and hopefully to identify potential biomarkers. I think where they are now is that in vitro experiments have identified a potential biomarker, which I believe is a pure(?) compound that has been shown to inhibit C. diff growth in vitro, and future experiments will look at the mouse model to examine the effects in vivo.

NCTR also has expertise in lipidomics analysis, which drove the collaboration between Dr. Akkoyunlu and Dr. Sun and Dr. Berger. The collaboration is looking at the unresponsiveness of infants' immune systems to the pneumococcal vaccines so that they require four doses. CBER, Dr. Akkoyunlu identified a shift in neonatal macrophages to an anti-inflammatory phenotype, and this shift was also observed when neonatal sera were incubated with adult macrophages. This shift was also associated

with formation of lipid bodies. The hope is that using lipidomics to understand how the anti-inflammatory macrophage contributes to neonatal unresponsiveness can lead to development of more effective vaccine. Lipid analysis showed that after exposure to neonatal sera, there is an increase in free fatty acids and triacylglyceride concentrations in adult macrophages. Further studies are currently being done.

The next ongoing collaboration is between Dr. Merkel from CBER and Dr. Cao, Dr. Xiong, and Dr. Heflich from NCTR. While there's been a resurgence of pertussis in recent years, that is not associated with nonvaccinated individuals. The increase is in individuals who have been vaccinated. Dr. Merkel has shown in some previous studies in nonhuman primates that the reason is due to the vaccine not really being effective enough to protect against pertussis infection and colonization. It protects against disease, but the individuals were still able to transmit the bacteria. So there really is a need to develop a more effective vaccine to prevent transmission and colonization.

So NCTR has this unique expertise in this air-liquid interface human airway tissue model, which is important for CBER because it's an alternative preclinical model for studying pertussis. The model could provide evidence to support regulatory review of vaccines, help

evaluate vaccine efficacy. It could also address the need for correlates of protection.

To date, they've demonstrated a stable infection of the air-liquid interface cultured with pertussis. And they will work on optimizing conditions to allow growth of pertussis to a high density following inoculation of the low-density, and also evaluating the contribution of virulence factors, identified from the nonhuman primate model through the infection of the air-liquid interface culture.

The next two projects involve genomic evaluation. First is the collaboration between Dr. Ye and Dr. Revollo from NCTR. They're looking at new methods to detect off-target mutations from genetic therapies like CRISPR-mediated genome engineering. The reason this is important is because these unintended mutations from these therapies are a major concern. They can cause cancer, they can cause other major problems, so there really is a need to develop ways to detect them. CBER collaborated with NCTR because of Dr. Revollo's expertise in developing these genotoxicity assays and NGS analysis. The outcomes of this study will help address a significant regulatory challenge in evaluating the safety of the technologies as they apply to the development of these advanced types of therapies.

In the past year they've identified an effect from a base editor that is independent of CRISPR-Cas activity. So as you can imagine this can have a significant regulatory implication, so further studies are underway.

The last ongoing collaboration is a project that was initiated by NCTR by Dr. Nakamura, who is working Dr. Sung in CBER because of her expertise in microfluidics. Current reproductive testing used in animals to examine the effects of fetal or postnatal exposure to drugs on spermatogenesis. This project is looking to develop and evaluate alternative preclinical models that use stem cells to replace the animal reproductive toxicological studies. Besides the impact of utilizing an in vitro test in lieu of animal studies, a microfluidic-based physiologically relevant tool for male reproductive toxicity would be innovative. I don't think it's been done, and would also allow for some interspecies comparison. The collaborators have optimized the protocol with the isolation of testicular germ cells from postnatal mice and differentiation of those cells on collagen-coated dishes. We're still working on the optimization of the germ-cell cultures in this microphysiological system.

The next few slides I'm going to briefly highlight some of the new NCTR-CBER collaborations that

began in FY20. Again, the majority of these collaborations focus on genomics-related studies for evaluation of in vitro alternatives. A new collaboration started between Dr. Ye and Dr. Revollo looking at the specificity and cellular effects of CRISPR-mediated inhibition of RNA viruses. These systems are being studied as potential antiviral therapeutics, so it's important to understand the impact on transcriptome integrity. The outcomes of this research could improve product safety and efficacy.

Continuing with the genomic-related studies, our next collaboration I believe is an extension of an ongoing large consortium, SEQC2. The next-gen sequencing provides a lot of information, as you know, and it can be used to diagnose different forms of cancer, generate tumor vaccines, et cetera, so it's really important to identify real mutations versus artifacts within sequencing. The one area that CBER regulates which has really grown in recent years is the use of NGS to generate patient-specific cancer vaccines that simulate an immune response against tumors. You can see we have some licensed approvals for some CAR-T cell therapies.

So there really is a need for development of reliable and robust methods to identify and understand the mutations identified from NGS. So this project involves evaluating the use of next-gen sequencing for product

characterization. Regulators need both lab and bioinformatic methods for the manufacture of personalized medicine, so that they can identify tumor-specific mutations and potential off-target effects. So the outcomes of this research could really help facilitate regulatory decision-making.

The next two projects are ones where NCTR is leveraging CBER expertise to support their projects. The first is a collaboration between Dr. Hart from NCTR and Dr. Stibitz from CBER, because of his expertise in live biotherapeutics. I think this also falls into the category of evaluating alternative in vitro models, because the investigators are looking to develop an in vitro vaginal tract model to assess live biotherapeutic products against toxin-producing strains of Staph aureus. If successful for CBER it could be helpful with strain selection and other aspects of designs of live biotherapeutic products to combat toxic shock syndrome.

The next project also focuses on alternative in vitro model systems. It's looking to evaluate countermeasures to prevent Zika virus sexual transmission. It's important because there really are no currently approved drugs or a vaccine to treat or prevent Zika virus infection. So this collaboration is between Dr. Petibone at NCTR and Dr. Rios from CBER, who is participating due to

her expertise in Zika virus virology. This alternative in vitro model is an in vitro cultured organoid that will allow us to study Zika infections, help us to identify means to prevent infections in immune-privileged testes, and could be evaluated as a tool for screening Zika virus antivirals for safety and efficacy.

Like I said earlier, in evaluating the collaborations between CBER researchers and NCTR scientists, we really found that most of our collaborations fell into these three areas, metabolomic and lipidomic analyses, evaluation of alternative in vitro systems, and genomic evaluations. We think collaborations in these areas have been really successful, and they've taken into account the strengths of CBER and NCTR, and we would really like to encourage additional collaborations in this area.

In summary, we found that CBER leverages NCTR expertise to develop methods and approaches to help support evaluation of our regulated products, but that NCTR also leverages CBER expertise to augment their development of alternative methods to evaluate toxicity of other FDA-regulated products.

However, like with anything, there are challenges to the collaborations. Collaboration science changes really quickly, the project may need to change with the science, the identification of synergistic opportunities

that are good for both CBER and NCTR continues to be challenging, but like I said before, we think we have some areas where collaboration can be formed or expanded. And not that this is specific to NCTR, but funding timelines and communications are always challenging issues.

With that being said, we've had some very successfully collaborations, and we look forward to continuing to work with each other in the future.

With that, I'm happy to take any questions.

DR. ASCHNER: Thank you, Emily. I will actually start with a question. Watching your presentation, it seems that most of these projects are already ongoing and sort of at some point the principal investigator recognizes that he or she might get some help from NCTR or vice versa, from CBER. I'm wondering if there's any sort of high-level discussions where the leadership of CBER and NCTR come together and they identify a project which can take advantage of both divisions.

DR. BRAUNSTEIN: I think we always try to encourage collaboration starting from the ground up, but I know that Carolyn Wilson, the associate director for research, she meets monthly with Donna Mendrick with NCTR, and if there are ever opportunities for collaboration, I'm sure that they're discussed and encouraged.

DR. ASCHNER: Thank you. Are there any other questions?

DR. TROPSHA: Just quick, I think there is a typo on slide 11, it should be A-L-I, not A-I model. Although you've talked about the use of AIs, so I think that's just confusion.

DR. BRAUNSTEIN: We will make sure we change that, thank you. I'll make a note of that.

DR. TROPSHA: Also, back to those models. I know that the types of models that are used currently in studying SARS-COVID-2. Is there any work planned in that direction, where you might use this model for either the vaccines or drug PKPD studies, for instance, or anything like this? Speaking of tissue models that you have, can those be used in SARS-COVID-2?

DR. BRAUNSTEIN: I really don't know for sure. I'd have to really check with the scientists. Not specific to those tissue models, but I think there are some projects that they are looking -- that they currently have ongoing, that they may be looking to expand to look at some of those SARS-CoV-2.

PARTICIPANT: Go ahead, Carolyn.

DR. WILSON: I just want to first thank Emily for doing a great job with the presentation, but just in terms of that last question, I think that right now, with COVID-

19, everybody is looking at all possible opportunities for gaining new scientific insights to understanding various aspects of the virology and the pathogenesis and how best to evaluate products. I think, in terms of using tissue models for evaluation of vaccines, I think the specific question, the challenge there is you would need to make sure that the tissue model incorporated all the appropriate immune effector cells in order to really be able to identify immune correlates of protection. That's our biggest issue with regard to vaccine development, typically. I think it may be more useful for some of the localized pathogenesis in lung or heart tissues, as opposed to vaccine development, but I also could be wrong. I think everything's on the table for looking at. I hope that answers your question.

DR. ASCHNER: Thank you. There's another question from an SAB member, from Mary Ellen. The question is, given the cutting-edge science in this space, how can NCTR and CBER scientists work together to further education of scientists in both groups?

DR. BRAUNSTEIN: We're always looking at ways that we can expand collaborations and make our scientists aware of what's going with NCTR and vice versa. We did receive a nice list of expertise at NCTR that we have been circulating to some of our scientists so that they become

more aware of the cutting-edge technologies and the expertise that is within NCTR.

We have a CBER scientist impact seminar which talks about at a high level the kinds of projects that we have going on, and if NCTR would videocast a similar seminar series, that could be something great where it would help facilitate the understanding of the types of research that are going on within both groups.

DR. ASCHNER: Thank you very much, Emily. Thank you for the review of CBER and interactions with the NCTR.

Let me go to my agenda here, and the next speaker is going to be Dr. Sruthi King and she'll be followed by Bernard Marasa, and they come to us from CDER. The Center of Drug Evaluation and Research.

Sruthi, please go ahead.

DR. KING: Good morning. Thank you for giving me the opportunity to talk and present today at the SAB meeting. My name is Sruthi King. I'm one of the associate directors of Pharm/Tox, in the Office of Generic Drugs, Division of Clinical Review.

Today, I will be presenting alongside with Bernard Marasa from the Office of Pharmaceutical Quality. I'll be starting off the presentation by talking about CDER's research goals. Given the short amount of time that we have today between the two speakers, I'll be going

through my slides relatively quickly. However, if you do have follow-up questions, please use the Q&A session or contact me afterward.

I will be presenting the CDER Pharm/Tox perspective and highlighting the type of work that we do, our current collaborations with NCTR, and identify areas where we could benefit from NCTR's scientific expertise to facilitate and further our regulatory review work. Then I will pass it on to Bernard, who will discuss the OPQ perspective.

Here are CDER's research goals. I won't read through everything because it's written on the slide, but what I wanted to highlight is that CDER's research enables us, in our regulatory review work, in some way, shape, or form, it does impact what we do, the goals regarding improving scientific approaches for pre- and post-market safety and efficacy, improving product manufacturing, testing, and surveillance. All of these research goals benefit the regulatory review in some way, shape, or form.

As I mentioned, I will start by providing the Pharm/Tox perspective, the OGD Pharm/Tox perspective and our collaborations with NCTR. Shown here is CDER's organizational chart, and the reason I wanted to mention this is because Pharm/Tox, or pharmacology and toxicology, reviewers are present in multiple super-offices across

CDER. I am located in the Office of Generic Drugs, which is on the bottom right-hand corner here, and we frequently collaborate with pharm/tox reviewers in the Office of Translational Sciences, the Office of New Drugs, the Office of Compliance, and we collaborate with chemists in the Office of Pharmaceutical Quality when making our safety assessments. Pharm/Tox is a consult discipline, depending on the super-office in which it is located.

So what does a pharm/tox reviewer in CDER do? CDER pharm/tox reviewer conducts safety assessments on INDs, which are investigational new drug applications, new drug applications, biologic license applications, drug master files, and ANDAs, which are abbreviated new drug applications, which I will discuss further.

Pharm/tox review informs the risk-benefit assessment, and we conduct a context-specific review, which means that we consider the dose, duration of use of the drug product, the patient population, and the route of administration in our safety assessment. As a discipline, we are unified in the approaches that we use to assess the safety of APIs, or active pharmaceutical ingredients, and other elements of the formulation, which include impurities, excipients, residual solvents, and extractable leachables from the container closure of the drug product.

We apply ICH and FDA guidances, and we use similar review tools in our reviews, and some of these are because of the hard work of NCTR experts, such as FDALabel and Smart Template. We also use an inactive ingredient database, among many other tools. Some of these are publicly available, so the FDA reviewer uses it alongside of industry, and really this facilitates drug development and transparency to industry.

We play a key role in establishing new policies and identify areas for improvement and review processes and tools that assessments can use, and support CDER's research activities, as necessary.

I mentioned that I'm part of the Office of Generic Drugs. Generics are submitted as part of the abbreviated new drug application pathway, or ANDA pathway. Generics have the same active ingredient, strength and dosage form, route of administration, and conditions of use. Generics are able to have differences between the brand drug and the generic. These include differences in excipients, or impurities, organic or elemental impurities that are inherent to the manufacturing process, or the formulation.

The generic must demonstrate bioequivalence to the reference listed drug, and because it is an abbreviated new drug application, a generic drug application does not

include the large clinical safety studies that are typically submitted to NDAs. Therefore, a generic relies on the prior evidence of safety from the RLD, and the information submitted to an ANDA should support that the generic has a similar safety profile as the RLD.

So our role as a pharm/tox reviewer in generics is also wide-ranging. The pharm/tox in generics is a consult basis type of review. So we do not touch every ANDA that comes through. The pharm/tox reviewer in the Office of New Drugs plays a role in every new drug application that comes through. The volume of ANDAs compared to NDAs is significantly higher, so pharm/tox is consulted when there's a safety question that comes up in a generic drug application.

We review pending ANDAs and also marketed products on a consult basis. Pharm/tox operates to fulfill OGD's mission to bring safe and high-quality generics to the American public. We assess the safety of the formulation of all aspects of the formulation, excluding the API. And safety assessment of marketed products is done under specific circumstances.

For instance, if there's a contamination issue of, for example, or a probable human carcinogen, or impurity limits that are out of specification, pharm/tox may be consulted to evaluate the impact of that

contamination or out-of-specification impurity limit. These types of safety assessments do have implications on product recalls and drug supply. If the contamination is found to impact a specific class of drugs and it's unable to be removed or mitigated, the product may be recalled to protect patient safety.

Alongside of that is doing the risk assessment, the risk-benefit assessment that I mentioned, to maintain patient access while the contamination issue is resolved.

Generic drug review is made possible by the Generic Drug User Fee Act. GDUFA enables us to have a predictable review process for generics. GDUFA also funds research to support the development of complex generics, where bioequivalence establishment is challenging. I've highlighted here the 2020 generic drug priorities and projects, and the goal of the GDUFA-funded research program is to improve drug development and bring more high-quality generics to the American public. The Office of Research and Standards within the Office of Generic Drugs leads the effort on GDUFA-funded research projects, and they collaborate with experts within OGD and across FDA, and pharm/tox is involved, depending on the project, on certain discussion or project discussions with investigators to help further that research.

Currently, OGD pharm/tox collaborates with NCTR on several projects. One of those is the FDALabel tool. This is a publicly accessible tool as you've heard earlier today, and as I think you'll hear further about later on today. It is used by FDA and also by industry. We benefit from the FDA labeling tool because it offers a full text search feature, and this allows us to conduct our context-specific safety assessments of excipients in generic formulations.

We have an ongoing collaboration for FDALabel enhancements that will further improve OGD's access to information in drug labels. This is a very useful tool for OGD pharm/tox, and several offices within OGD, such as Division of Labeling Review, and we benefit from this collaboration.

Additionally, a second collaboration that is a little bit earlier in its phase is the Smart Template, which is currently being used by the Office of New Drugs for IND reviews, and this is something that we are working with NCTR even as recently as last week, we had a meeting with NCTR collaborators to adapt this template for OGD pharm/tox review, and perhaps bioequivalence review. Review tools such as these would help us improve data capture from reviews, collate and centralize toxicology

safety information to allow for improved data mining and analysis.

On the next slide, I would like to highlight some areas where we could benefit from the toxicology expertise at NCTR. As a pharm/tox reviewer of NDAs and ANDAs, the types of studies that are submitted for review include pharmacology, general toxicity, reproductive toxicity, in silico predictions, genotoxicity, and carcinogenicity. In silico predictions are something that we see with increasing frequency in generic drug review, and these are submitted for genotoxicity assessments, for carcinogenicity predictions, and also when empirical data are absent, for chemicals that are identified as impurities or contaminants in the formulations. Surrogate analysis and read-across approaches are increasingly becoming common in an applicant justification. ANDAs may include de novo genotoxicity and general toxicity studies. However, there's many gaps that we have identified as regulatory reviewers.

Applicants may not always be familiar with the guidances or the best approaches for safety justifications. They are limited in the extent of studies that they are willing to conduct, and as generic drug reviewers, we are limited in the extent of studies that we can request from generic applicants. For example, if there is a chronically used product that has a missing safety justification for

chronic exposure or potential genotoxicant that needs evaluation of carcinogenicity, the applicant would be referred to the Office of New Drugs as it cannot be submitted as a generic application.

There are also data integrity issues that we encounter. This is because we have CROs, contract research organizations, that are not always familiar with the rigor with which the studies need to be conducted. The data integrity issues further complicate the regulatory review process as we need to coordinate inspections of CROs, many of which are located outside the United States, and one CRO may submit data or may provide studies to support multiple ANDAs. Therefore, data integrity issues also impact the knowledge gap that exists in data necessary to make informed regulatory recommendations.

What are some of the ways in which we can collaborate with NCTR tox experts to bridge this gap? Relying on NCTR's expertise to conduct and optimize studies for specific compounds or compound classes for which relevant safety information is sparse or nonexistent would be very beneficial for the regulatory pharm/tox reviewer, both in OGD and the NDA side. This is because when doing reviews we see similar compounds or similar compound classes coming up in reviews and we struggle with what is the best recommendation for the applicant to resolve or

remove the safety concern. So relying on NCTR's expertise would facilitate better risk assessment and improve consistency in the recommendations that we can provide to industry for ANDAs and NDAs.

This was a short presentation, but what I hope I've highlighted for you is that pharm/tox does review a wide variety of studies and applications. We benefit from the collaborations that we have ongoing with NCTR. And there are areas in which NCTR's expertise in toxicology, along with many other areas of expertise, can help us bridge the gap in knowledge that exists between what we have available and what applicants submit, and bridging this gap would actually benefit both ANDA reviewers and NDA reviewers in the pharm/tox discipline.

Now I'm going to hand over the presentation to Bernard, who will present the OPQ perspective. Thank you.

DR. MARASA: Good morning, everyone. My name is Dr. Bernard Marasa. I am a review microbiologist within the Office of Pharmaceutical Manufacturing Assessments here in CDER.

Prior to coming to CDER, I first must thank NCTR for the training. I was one of the FDA commissioner fellows who trained through NCTR, with the leadership of Dr. Saeed Khan and Dr. Cerniglia. From there I was able to transition to my current job as a micro reviewer within

OPQ, within the Office of Pharmaceutical Manufacturing Assessment within the Division of Microbiology Assessment.

Today, I'll be sharing with you the details of an ongoing project that when I joined the center, I was able to continue as the point of contact, and that is the collaborative projects within CDER and NCTR that involve detection of BCC in pharmaceutical products regulated by FDA.

Quick, with respect to Office of Pharmaceutical Quality, within CDER, which was formed in January 2015, and it's mainly responsible for quality review functions for new drug applications, generics, and biologics. So it basically integrates quality review functions with inspections, surveillance, policy, and research. It's a large, team-based review.

Within OPQ, there is the Office of Pharmaceutical Manufacturing Assessment, or OPMA, and its mission is to assure that quality pharmaceuticals are consistently manufactured over the product lifecycle. And the vision basically is to be a premier regulatory organization for holistic assessment of pharmaceutical manufacturing. Some of the key roles include serving as the OPQ's centralized resource on pre- and post-approval inspections and manufacturing issues that may impact application assessments.

Within OPMA, we have the Division of Microbiology Assessment, and this basically serves as OPMA's centralized resource for expertise on microbial control, sterility assurance, microbial product quality aspects for pharmaceutical manufacturing of small molecule drugs, and it's also involved in reviewing NDAs, BLAs, ANDAs, supplements, INDs, drug master files, and meeting packages.

So, we mainly review microbial controls also of nonsterile drug products, and this is where the topic of today's discussion falls into place and BCC actually mainly can find in an unsterile aqueous drug products that are listed by the FDA.

What exactly is BCC, or Burkholderia cepacia complex? It's composed of at least 17 or so similar but genetically different species, and the most common that are being identified include cepacia, multivorans, and cenocepacia.

These are complex organisms that are gram-negative, and they are very remarkable for their adaptability and resistance to a large variety of antibiotics and microbial agents. They are mainly found in a lot of ecological niches including the plants, soil, and river water. And as you may be aware, water forms a huge part of a lot of pharmaceutical products, so it's a big concern that this microorganism must be controlled.

So FDA is very concerned with presence of BCC in pharmaceutical products and its transmission especially to immunocompromised individuals. There have been a lot of contaminations and recalls in the past, and the investigations have always led to BCC, with a lot of field alert reports, drug recalls eventually, and there have been several deaths of Americans. So there's a huge concern for the agency to make sure there are risk mitigation strategies to control BCC in pharmaceutical products.

In 2011, the agency came with a publication mainly trying to provide guidance to industry on how to mitigate issues of BCC contamination and especially in nonsterile aqueous drug products, and with that led the Division of Microbiology Assessment to actually generate what we call the boilerplate language which we could provide to industry applicants whenever they submit an application, and they don't provide any BCC risk mitigation strategy with that, we tell them we need you to show that you're able to show that there's no potential contamination of BCC in the products you are using to manufacture your product.

With that, there are several Code of Federal Regulations that maintain safety for the products that are being produced, especially on microbial contamination. Section 211.113, appropriate written procedures, designed

to prevent objectionable microorganisms like BCC in drug products are required to be established and followed.

Another one, 211.165, there should be appropriate laboratory testing, as necessary, of each batch of drug products required to be free of objectionable microorganisms.

And then USP <1111>, one of the oldest, requires companies to either use either USP 61 or 62 to show that they have control of these objectionable microorganisms in their drug products.

The current collaboration with NCTR, the need was, of course, to develop methods that can help detect and characterize microbial contaminants in FDA-regulated products, and the impact of this collaboration was to reduce drug recalls due to microbial contamination and enhance approval of safe drugs for the American public.

I'll say this collaboration has been very fruitful, and as the agency we are extremely grateful for the great work that has been done by our collaborators at NCTR, especially Dr. Ahn and Dr. Carl Cerniglia. The current results from the collaborations show that they were able to show that they developed comparative methods to study the detection of BCC in pharmaceutical products, and also over the years, they have been able to develop a

resuscitative step and enrichment technique for BCC recovery.

They also developed, demonstrated that diluted TSA and TSB media and R2A and R2AB media showed better recovery and efficiency than TSA and TSB. They were able to demonstrate that some of the BCC strains could remain viable in antiseptic for even up to 28 days. And some of their work through this collaborative work, they were able to actually develop a rapid PCR-based detection method for BCC from water and other pharmaceutical drug manufacturing raw materials. I must say that this collaboration has been very productive, and because of the large body of data from the collaborative work with NCTR, the PDA committee, the microbiology committee, caused from the (indiscernible) that they were able to recommend a test method for BCC detection.

For a long time, industry was mainly using USP 61 and 62 and USP 1111, but as I said early on, those methods are not very specific for BCC detection. But from the work at NCTR, in connection with other regional labs, the microbiology committee in the third year, they were able to convene the USP to actually develop the USP 60 now which went live in December 2019, that is very specific for BCC detection, and this has made the work of micro reviewers, especially in our division, to be very simple, because once

an application comes in and we don't see use of this method to show that the product is safe, then we refer the company to make sure that they are able to show that they are able to detect these microorganisms using this method.

One of the big concerns in a lot of meetings with industry was there wasn't a specific method for detection of BCC. But with USP 60 now, courtesy of the work done by Dr. Ahn and other regional labs, we say that this problem was solved.

I actually wanted to give kudos to Dr. David Carson, who started this collaboration, and last year, 2019, PDA meeting, he was able to actually give credit to the work, the collaborative at NCTR, with the publication they did for the knowledge that they gave to the industry that led to adoption of this USP 60 that went live in December 2019.

There is still current collaboration with NCTR even after the adoption of USP 60 for detection of BCC. Last year still we felt we need to have a comparative study for any other methods that are out there, and Dr. Ahn and the team at NCTR are working on this, and this is the current collaborative project that is ongoing.

In conclusion, I'll say the collaborative research on BCC detection has provided data that has contributed to the USP publication of a test method for BCC

detection that wasn't available until December 2019, and this is USP 60. A very big achievement for collaborative projects. And currently the ongoing collaborative project that seeks to enhance sensitivity of BCC testing by developing comparative methods for its detection hope to improve farther on this work.

To conclude, I want to give thanks to collaborative partners at NCTR led by Dr. Cerniglia and OPMA for BCC research funding and support.

Thank you.

DR. ASCHNER: Thank you, Dr. King and Dr. Marasa. We do have one question from Dr. Chris Whitehouse, and the question is are there any microorganisms in drug products that are not objectionable? One of you please answer it.

DR. MARASA: Yes, very good question. When we are reviewing the stability of an application, before they (audio interference) that are submitted that are under (indiscernible comment). So these are allowed to have microorganisms, but their level is controlled. So the tests are there that are done and comparative methods that the applicants need to show that they are run through, and their products fall within those test margins. So they use comparative methods with USP 60, for instance, what I was just talking about, just BCC detection, they need to show that if their products (indiscernible word), they need to

show that they control for BCC detection. They also need to show for control for USP 61, that's the enumeration, microbial enumeration. They need to show that the number of microorganisms within there fall within certain margin, which is controlled under USP 61. And those are the objectionable microorganisms, like Pseudomonas aeruginosa), BCC that are controlled under USP 62. But for that list that are regulated by FDA, of course, no microorganisms are expected to be found there. So they use USP 71, which is the stability(?) testing for the release of their drug products.

DR. ASCHNER: Thank you. Are there any other comments or questions? We have time, so please if you have any questions, go ahead. This is for the SAB members, so I should make it clear, sorry.

(No response.)

Okay, if not, we're going to move on. Thank you very much, Dr. King and Dr. Marasa.

Our next speaker is ready, is Dr. Ed Margerrison, from the Center for Devices and Radiological Health. Please go ahead.

DR. MARGERRISON: Thank you, and good morning and good afternoon, everybody. My name is Ed Margerrison. I am the director of the Office of Science and Engineering Labs at CDRH. I know a lot of people have talked about the

organization within their different centers. I am extremely fortunate because essentially all of the research that we do at CDRH comes under one office, which it organizationally significantly more simple, I think, than it can be in some of the other centers.

I've used the next couple of slides before, but I wanted to just give a little bit of an overview again of CDRH. Many of my colleagues who are on the phone always laugh at me because I always say that CDRH is a little different from the other centers, and they all chip in and say, well, we all are. But we are a little different from certainly the other human health centers because I think the type of product that we're dealing with has an incredibly broad technology base, and clearly that's true of all the centers.

We deal with currently 190,000 types of products. We're getting close to three-quarters of a million individual products that are on the U.S. market, and we cover three types of products, broadly. There's medical devices, which I'm going to talk a little bit more about and what they really are, because they, and I know I'm repeating myself from last year, but I think it is worth the repetition. We also cover in vitro diagnostics, which, as you can imagine, has been an interesting few months in

our center, trying to keep on top of what's been going on there.

We also deal with radiation emitting products. These are essentially anything that's a consumer product that can emit radiofrequency radiation. In the vast majority of cases, we don't have any sort of premarket regulatory mandate over those, but certainly as it relates to public health and safety, we have the legal requirement to recommend to other bodies, for example, FCC, as to whether we think there is a safety issue there.

Like the other centers, we have in most cases pre-market regulatory authority. We're also responsible for the manufacturing facilities and quality systems therein, and of course post-market safety of many of those products.

I'm going to come back again to what actually is a medical device? We frequently joke that if you go into a healthcare clinic or a hospital, the first hundred things you see are probably going to be medical devices. It is a thing, a machine, a contrivance; nowadays it also includes software, of course, that is intended for use in diagnostics of disease or in treatment of a condition. But crucially, where we're very different from our friends and colleagues in CBER and CDER is that our products

specifically exclude a chemical effects on the body that is not intended.

So we're very much dealing more with very often the engineering side of life sciences. The very common medical devices that people know and love are things like total hip, total knee replacements, pacemakers, stents, and a variety of other devices, as well. I could go on. But I would bore you rigid.

So that just gives you an idea of the breadth of what we're dealing with. As an organization, OSEL, as we call it, the Office of Science and Engineering Labs, that conducts most of the research, we get involved in about 25 to 30 percent of the premarket device reviews across the center, of which there are somewhere around 22,000 to 25,000 per year.

We get approximately 1.5 million field reports on medical device events and malfunctions, just in the United States. And because, as I said, we're responsible for the medical device facilities, it's worth noting that there's about 25,000 of those, which belong to about 18,000 different medical device manufacturers in the United States and across the world.

That's just a bit of an introduction to give an idea of what our world is. Coming back into the research, what I want to do is give you a bit of a feel, rather than

talk about specific collaborations, to talk about an initiative that we've been doing in the organization to actually do something very different from what Emily was talking about a few minutes ago. We are moving our research to being instead of opportunistic, individual collaborations, to being much more of a top-down, strategic approach, and I wanted to talk about that, and then talk a little bit about some of the huge issues that we're currently facing as a center.

We have reorganized ourselves over the last 12 months or so into approximately 20 programs. These, in many cases, do parallel our pre-market offices. So for example, we have a pre-market office of cardiovascular, we do have a cardiovascular program. We have an orthopedics program. And some of the others that are more technology-based are here up in front of you. The largest single research program that we have as an organization is artificial intelligence and machine learning.

I noticed that Adm. Hinton did call out Donna Mendrick for being the chair of the agency-level workgroup on AI, and we've had an enormous amount of help from Donna. Thank you, Donna, in that area. And it's been an area that we've been driving as a center for many years. AI machine learning is not new to us. We cleared the first device which was actually a diagnostic device for pap smears, that

uses something along the lines of AI, in I believe 1997. That really highlights where, as a center, we certainly are different as it relates to AI and machine learning, because the whole world can learn an enormous amount about how to do things better from an infrastructure perspective using AI and machine learning. We actually have devices that are based on AI and machine learning, and actually have adapted algorithms and things like that built into them already.

It's achieving a huge amount of use in diagnosis of disease, particularly around imaging and visualization, and we're beginning to really learn an awful lot more about its use in digital pathology, with some collaborators at MGH and Harvard. That's an area that, it won't surprise anybody on the phone to know, is going to get bigger and bigger.

I wanted to highlight, as well, I will talk a little bit more about what we've been doing in the emergency preparedness area. This is actually a new program for us that we're pulling together at the moment. It obviously has its origins in the terrible events of the last four to five months, and we are really now just beginning to kick this off. Not from a perspective of what are we doing right now. We have a number of research projects in this area ongoing. But really about what do we need to do much more strategically in the medium and long

term to try and actually get ourselves a little more ready for the next time. That's going to be massively important for everybody.

So we're continuing our short-term efforts and some of those involve, of course, the channeling of many of our staff into doing more premarket reviews. If you would care to have a quick look at the FDA external website, there is a page devoted to CDRH and a lot of the work that we've been doing as a center during the pandemic, and a lot of the emergency use applications, pre-EUAs, all of the diagnostic clearances, and the approvals in some cases as well, have been coming through us. It's been a time of literally all hands to the pump. But it's been quite rewarding in a rather weird way, as well.

A lot of what we've been doing in that area has been very public. We've been right in the middle of the PPE shortages and trying to put together things like companies who have expressed that they've got actually capacity that's spare, what can they do to help? We've been putting them together with particular people to try and help make PPE and several of those things.

That gives you an idea of some of the things broadly we've been doing from a research interest, and I'm going to drill down a little bit and give you some more specifics, and then talk about what we're doing.

Within our current 20 or so research programs, we are currently putting together charters, and this is very much deliberate for what I was just saying, to try and get a lot more of a top-down approach. I'm lucky enough to have a staff in my office who are phenomenal, great expertise, great experience, great creativity, and some phenomenal scientists and engineers. What we're trying to do is to get a bit more of an overall prioritization for some of the work that we're doing.

This has been a bit of an ongoing event for a little while. The current phase that we're doing is that we're very close to being at the end of defining what we're calling our project charters or our program charters, and the reason for doing this is something that I have mentioned to the Scientific Advisory Board before. The breadth of technology that we're seeing in CDRH is going up in a linear fashion.

That means that, being absolutely blunt, we've got to be very, very good at prioritizing, and we've also got to be very good about trying to get other organizations to do work which will be on our agenda. Otherwise, we're going to get to the stage where we can't cover all of the technologies, and we're beginning to get there right now. And if that happens, then we're going to get manufacturers and sponsors coming in to get their devices and their

diagnostic devices cleared or approved, but we do not have the bandwidth to even know what are the appropriate questions to ask them so that we can be comfortable to understand the fundamental safety and effectiveness of those devices, which is what we're trying to do.

So we need to be very, very focused, and that's what we're currently doing. I've also recently employed something which is very -- a person who is a great help to me, but very different for a typical government position -- I actually now have a business development director, who is going to be right in the middle of all this and actually working very, very closely with the National Science Foundation, the National Institutes for Health, to really start making sure that we have a common agenda which will be based on the priorities that we're defining. So that's an ongoing exercise at the moment.

One of the things that we're also doing in this area is that we will be publishing these, and by publishing I mean putting them on our websites, and then working with various organizations; for example, the Association for the Advancement of Medical Instrument, or AIMI, is an organization that we work very, very closely with, and they are helping us to publicize some of these things. They'll go on our website so that it's crystal clear to the outside world what our priorities are.

That hasn't been crystal clear up to this point, because typically our research portfolio has simply been the summation of the individual projects that are going on. So we're not looking necessarily to make wholesale changes in those projects, but that we will be seeking very, very specific collaborations in these areas and not just waiting for collaborations to happen. They're going to be much more directed by our strategic framework.

So that is an ongoing example, as I've said. I wanted to give you an idea of how we're really trying to focus in on some of these things. Additive manufacturing is again an area that to CDRH is about three decades old. We have a number of areas, number of products that we've cleared and approved that are made through additive manufacturing or 3D printing. So in some ways, we're very much at an inflection point of what regulatory science needs to be done with additive manufacturing. So redoing a project charter at this point or a program charter at this point in time makes a lot of sense.

We have issued guidance which is, if you're not aware how the FDA makes its policies public, to give industry an idea of exactly what is required, what's needed to get clearance, or approval of a device, and in the case of additive manufacturing, we've issued guidance which really specifies some of the technical consideration that

are required if you want to put an additive manufactured product onto the market in the United States.

But times, they are a-changing, without a doubt. What we're now trying to do is to say, well, what are the next questions coming up? The first two very much are an extension of those technical considerations. We want to try to make sure that the additive manufacturing processes that are used by industry are crystal clear and that the validation process for those is very transparent, very consistent, because that will help the whole of the med tech community.

We also need to make sure -- and this is something that is never going to finish, but we need to make sure that the testing is appropriate for all of those additive manufacture devices. We need to make sure that the software is appropriately validated. This is really a way that we're trying to really get ahead of the curve, because as an ex-medical device manufacturer myself, one of the things that I would love to do would be to sell software rather than the actual device. I would love to push that responsibility onto other people, because it would be so much more efficient as a device manufacturer.

That really leads on to the third huge goal and gap that we're trying to start getting our heads around. We know that 3D printing of devices at the point of care,

i.e. within healthcare facilities, is something that has started, and it will continue. We're starting to really try to start creating a framework for allowing people to do that.

Given the way that our intention within the med tech community is to spur innovation, the last thing that we want to do at the center is to dampen the enthusiasm for this, because the advantages for patients are immense. Being able to get really personalized devices 3D printed at the point of care is potentially significantly better from an effectiveness of the device perspective, and also potentially significantly quicker to actually get a device to the patient. So we're very, very keen on doing this.

But there are a lot of regulatory questions that are being raised. We don't have the answers to a lot of those questions, but we're beginning to have those discussions with healthcare facilities, with healthcare professionals, and of course with industry to try to work out how we're going to do this together. So that's a great example of how even though the actual 3D printing medical devices, I think the first one was cleared probably 15 or 20 years ago, but the whole area of this technology is evolving, and that's very, very typical of what we see in the medical device community that the vast majority of devices really do evolve from previous devices. The

technology broadens and expands and becomes new, but we don't quite as often I don't think see something as radically new as something like CRISPR-Cas9 in the device industry as our friends and colleagues do in some of the other centers.

So that gives you an example of what we're trying to do, I think, hopefully with the project charters. I wanted to also raise something that actually became public yesterday. So this is tremendously good timing. We have spent a lot of time doing some phenomenal research that gets published and not enough people are aware of it. The story behind those publications and the real regulatory science implication for patients, advocacy groups, and healthcare professionals really remains undiscovered, because too many of those publications just gather dust.

So one of the things that we're doing as a center, as I said, this started yesterday, is that when we have developed tools that can be useful across the whole of the med tech industry, then we're going to publish those in a catalogue on our public facing website. I don't actually have the URL yet. So I was going to put it on the slide, but I will find it and send it through, Donna, if that's useful.

But we're starting to detail in different categories things that med techs, particularly the small

companies, may find useful, and from my perspective, having worked in a small number of small med tech companies, this is massively important for them. We see in the device space that the vast majority of innovation comes through small companies, not through the large multinational global companies, but the small ones. They're really the people that start driving innovation, driving new technology, and frankly, having worked in them, they don't have time to read journals and scientific publications. So this is our attempt to try to allow them to access things that are going to help that early stage of the transition between technology development into product development. That's the most risky time in device development.

One of the things that we want to do is to provide methods, tools, for example, virtual and physical phantoms that can be used for scanning and things like that. We're making the availability of those much, much more public so that people don't have to know where to look other than in our science catalogue. We're also publishing a lot of the methods, which may not be something along the lines of a medical device development tool exactly, but if it's a laboratory method that's been developed within CDRH, it at least has some stamp of regulatory authority and within an appropriate use can be very, very useful to people.

Fundamentally what we want to do is to allow the small med tech startup companies to concentrate on the technology development so we know how good their product is and we can then -- or have to have less questions about how good their testing of those devices is.

That's one of the things that we're trying to do with our catalogue. I would very much love it if you were to all go and have a look. It will push our hit rate up.

A few words about what we've been doing in emergency preparedness and what we intend to do. Clearly as I mentioned earlier, this is the result of what we've all been through the last few months. Some of the things that we've already been doing are we've been looking at the airflow around masks of various types, everything from full respirators through to homemade cloth masks, and we've been doing some computer modeling and simulation about how good those are. What different materials are good for making masks at home or in an industrial setting, what we can expect them to actually do from being able to filter out particles of different size, et cetera.

That's an area that we want to massively expand, particularly as it relates to masks that can be used for children and other people because the number of masks that are actually publicly available specifically designed for adolescents and children is very, very low.

So one of the things that we're trying to do is again it's an example of how we're trying to stimulate innovation by making models publicly available for predicting airflow and therefore effectiveness of masks, depending on the different materials and designs, et cetera, we want to stimulate some innovation in that space, because innovation in masks has been notoriously slow for a number of years. And of course the urgency has gone up massively and very, very quickly, and we've been very fortunate to be able to continue this bit of work even though our labs have been largely closed during the pandemic.

Hugely important area for us as always is in developing new testing methodology for things, and we have been looking at the current typically used testing methodologies for PPE. For example, gowns. Typical testing methodology does not account for the use of things like gowns and full hazmat suits under real world conditions. This became apparent when we started during the Ebola outbreak in Congo, and it's something that's becoming more and more important.

One of the things I just wanted to highlight as well just from the question of emergency preparedness we are not involved in diagnostic development at all. That would be a conflict of interest for us, because we cannot

develop things that we subsequently regulate. But we have been very much intimately involved in the diagnostic part of Project Warp Speed to make sure that the data once it comes into CDRH is what's needed for us to be able to clear devices.

So I want to talk about where we're going in the future. I've talked about how we're at CDRH we're setting ourselves up to redefine really what our program areas are. One of the key things I think that is a very, very important area for potential collaboration between ourselves is there's a number of materials where we are beginning to be much more aware that there may be long-term risks to those, and so I'll leave this slide up and just say what are those materials. We really want to start understanding that. We have had a very public workshop looking at the potential effect, long-term effects, of things like dental amalgam, metal ions, this is an area that is going to become bigger and bigger and bigger in the future without a doubt.

So as I've said, the things that we're really interested in, and I'd challenge the whole med tech community to, as we publish more of our specific aims, is to really look at how we can apply research across the whole of the community to help us, to the big things that we're looking at, how do we stimulate innovation in those

medical devices at an early stage to keep technologies alive for longer? And then the fundamental question in devices, what really happens to those materials which are in the body, because many of those are left in the body permanently as permanent implants. We need to know a lot more about that.

So on that, I will close and welcome any questions or comments that people have.

DR. ASCHNER: Thank you, Ed, for a very interesting and enlightening in many ways presentation. Obviously with the pandemic, you guys have been very busy. The one thing that I didn't hear from you is, since we are reviewing the NCTR, is what are the interactions between CDRH and the NCTR?

DR. MARGERRISON: There's actually an awful lot of them at a very granular level right now. One of the areas we'll probably hear more about over the next day or so is a lot of very, very close collaboration relating to the NanoCore and facilities. We have a sister facility in one of my buildings, which used to be called the nano core, is now called the Advanced Characterization Facility, the ACF. We have hugely close collaborations in those areas as well.

The other area I think that we heard a little bit about when we talked about the neuro area earlier is I remember the first of these meetings that I attended where

I highlighted the massive importance of tibial(?) brain injury in early TBI. That's another huge area of collaboration for us. So we have -- I can't remember, I'll be honest, the specific number of projects, but the collaborations are very much alive and well in a big way.

DR. ASCHNER: That's great, thank you. We have a couple of questions from SAB members. The first one is from John-Michael Sauer. The question is what will the CDRH/BD type collaborations with external stakeholders look like? Will these collaborations take the form of PPPs or BAA contracts?

DR. MARGERRISON: There may be BAA contracts, but the fundamental thing that I'm trying to do, because we have very little money, is I really like to -- and we're already working closely with a number of the partner institutes of NIH -- to really start getting them to recommend project areas that will be -- imagine the Venn diagram. We have a huge amount of overlap with NIH clearly, and also NSF.

One of the things that we're contemplating doing and I can't make this public yet -- this is a public meeting, but I could very much envisage a partnership with parts of NIH that would, for example, stimulate the development of new medical imaging phantoms. A great example of this is last year we published on the very first

official medical device development tool that was developed through my organization and has become available.

I was having a conversation with one part of the NIH, and they said, well, that's great, because this is in the area of high intensity therapeutic ultrasounds, and it turns out that they were currently funding six new SBIR companies in the area of high intensity therapeutic ultrasound. So if we can get together and get those companies using industrywide tools, then NIH can fund their companies to develop that technology, not to develop six different types of evaluation methodology, because that's completely inefficient for everybody. So essentially, we can drive innovation and efficiency at the same time. Does that help answer?

DR. ASCHNER: He said thank you, so I assume so. Thank you. We have another question from Patty Ganey. The question is with respect to the mask innovation for children, is this information provided in a manner that moms could make a better mask for their children, or is this only applicable to commercial development?

DR. MARGERRISON: That's a great question. We want to do both things, because we are not there yet. This is something that is an ongoing project. We have done some publication on good materials and there's some more publications coming out. But eventually absolutely. We

want to be able -- one of the things that we've been hit really hard with over the last few months is people want to know, well, I want to make a mask. What specifications are there? Clearly, as a regulatory body, we cannot publish specifications that companies have given to us. It's proprietary information.

So one of the things that we want to do, and again, we've been doing this very closely with NIH, for example, the 3D printing, the NIH print exchange is a collaboration between our 3D printing group and the NIH, which allows those generic specs to be public. So eventually those specs, for example, could be used by my wife in our kitchen making masks or by 3M or anybody -- that's not an advert for 3M -- or by any other medical device manufacturer that wants to make those as well.

Like many of these things, it's horses for courses. There will be a need for high-grade N95s, full respirators, et cetera, but we know that -- CDC say this all day every day -- there is a massive need for those homemade masks as well.

DR. ASCHNER: Thank you. Dr. Ganey says thank you, as well.

We have time. So Dr. Lanza has another question. I think he's typing it actually as we speak. So go ahead, Greg, type it please.

(Pause.)

Why don't you type it, because we can see what you're typing? Okay, so he is saying he'll skip it. You're off the hook, Ed.

DR. MENDRICK: This is Donna. We should break for lunch.

DR. ASCHNER: Okay. I don't see questions, we'll break for lunch. We're almost on schedule actually. So why don't we reconvene; I believe it's 1 o'clock Little Rock time. Yes, 1 o'clock Central time. It's going to be 2 o'clock Eastern time. I did the math. So it's 11 o'clock Pacific time, and it's going to be noon Mountain time. So we'll see you in about an hour, 55 minutes to be exact.

Thank you, everybody that presented this morning. And we'll resume at 1 o'clock Central time.

MICHAELKAWCZYNSKI: All right, yes. We are going to take a 55-minute break. So you can stay connected in, and we will see you, like I said, in about 55 minutes. Thank you much.

(Luncheon Break.)

AFTERNOON SESSION

Agenda Item: Public Session

FDA Center Perspectives (Continued)

MICHAELKAWCZYNSKI: Welcome back to the NCTR Science Advisory Board Meeting. I hope everybody had a wonderful lunch, and also, you know, I'd love to ask a question of, hey, what in your lunch was impacted by FDA, CFSAN, or any other agencies. But all right, that's for another time, another day.

All right, Michael, are you ready? All right. Take it away, sir.

DR. ASCHNER: Okay, we have a couple more presentations before we have the next break, and the first one is from CFSAN, and everybody can see Dr. Fitzpatrick already. So please go ahead.

DR. FITZPATRICK: Thank you very much for inviting me to talk about some of the exciting things that are going on with CFSAN in toxicology, and as we were talking earlier at the break with Michael and all, the products that we regulate touch every American every day. We regulate food, 90 percent of the food supply, food additives, food packaging, color additives, flavors, botanicals, dietary supplements, contaminants in food like arsenic and lead, cosmetics. A lot of the products that we regulate, we don't have any preapproval authority on. So we rely on

research that we conduct in our lab or in conjunction with NCTR to make sure that those products are safe, especially safe for the American public.

So today I'm going to talk about the CFSAN activities that we have going on, our partnerships, our research at OARSA which is our lab, and the CFSAN research that we do at NCTR.

So, you heard me talk a little bit about all the compounds that we work on, and one of the big things that we work on is flavors. So that was a big issue of flavors. There's so many different chemical flavors, we couldn't possibly have tested them all in animals.

So what happened in 1978, FEMA -- that's the flavor industry, not the people that help disasters -- designed this decision tree which divided the flavors -- it was originally used for flavors -- into three different toxic classes. And those flavors then, that determines how much toxicity testing you needed depending on the level of exposure, some of the criteria and classes that they were.

And that actually led to the development of what is called the threshold of toxicological or concern, or TTC, which originated in the foods part, but is now being used in other parts of the world or for other toxicology classes, and for which there's a lot of interest.

So briefly we discussed expanding the decision tree approach, and this is of great interest, I'm surprised, to the Europeans and everywhere, into a lot more, six classes versus three. We increased looking at the different functional modalities. We looked at mode of action, species differences, whatever we knew about it, and we approved this to get what looks like a very complicated expanded decision tree.

But once we finalized this and then we're publishing a series of papers on it, I think it will be a very effective tool to decide where we should be putting our resources for tox testing and where we don't have any concerns about that. As you can see, as most of our products, except for food and color additives, we have no preapproval authority over, so it's a real challenge for us to prioritize and make sure all these compounds that are found in the food, cosmetics, et cetera, are safe for the public.

Additionally, we wanted to look at whether, we wanted to reassess whether we needed a dog study for food and color additive safety assessment. Those are the two areas where we asked for a toxicological package in order to approve those for use in food. This was an assessment for food and color additive safety only. It doesn't apply to where a dog study might be used for a safety of another

compound that FDA regulates, but we wanted to look at the impact of the dog. We don't get that many food additive petitions anymore. Most things come in through GRAS, Generally Recognized as Safe, or GRAS. But we looked at 162 food and color additives from 1950 to 2018 and found really that for food and color additives, we didn't see any unique toxicities, and that possibly we could combine the rodent study with some ADME data in order to give us sufficient confidence in the safety of a food and color additive if it came in for an approval.

We also are very concerned about toxic elements in food. We call toxic elements metals. Our chemists call them toxic elements, and we're very concerned, and I know I've spent a lot of time starting with looking at levels of arsenic in children's food. So arsenic in children's, in infant rice cereal was very high.

But we go out routinely now, which has been kind of stopped with the pandemic, and do what's called a market basket survey, where we go and pick up from the grocery store in our regional offices different commodities, and then we look at them for what's in them, and we can see that not only is arsenic in infant rice cereal, infant products, but other metals: cadmium, lead, cadmium, lead, arsenic, and mercury.

And so we're not only concerned about those metals as individual, but more importantly how do they act in mixtures? This toxic element strategic framework is what Dr. Mayne, our center director, set up to really have a functional program to look at what metals and what mixtures of metals we should concentrate on.

And for this one, we have the management group, then we have -- then they give a question to the science group based on regulatory questions, and then it goes through a whole loop to make sure the questions are answered scientifically, and then it goes back through the Office of the Director and the deputies to make a decision, and then how to implement it.

But at the same time that we're implementing that plan, we also have a communications strategy to make sure that the public knows what we're doing and what the consequences of our action are. So that's been a really exciting program to be part of.

And because, as I said, we had so many products that we don't have preapproval authority over, which means we have to do some sort of research in order to show whether how that product gets used by the label instructions might be causing harm, alternatives are very important to us. Alternatives to animal testing are really important to CFSAN, and that's why -- one of the reasons

why first myself and Donna Mendrick and others worked on the predictive toxicology roadmap, where we emphasized that FDA was interested in alternatives, and then we created, Donna and I, created an alternative methods working group across all of the FDA. So this has members from each of the centers that you've heard today, senior members from each of the centers that you're hearing from today, as well as NCTR and our field offices and the Office of the Commissioner.

To strengthen FDA's commitment to promoting and developing these new predictive toxicology technologies to reduce animal testing and to make sure we're discussing as an agency, and very excitingly, just a couple of days ago, the commissioner tweeted about our alternative methods working group and asked people to go to our website, which is the website is visible, this is a public website, where we are committed to putting updates on things we are doing for alternatives, so that the public can really see.

Some of these studies, things that FDA are doing, and we are doing a lot of exciting things in this area. All the centers are. One of the first things we're looking at, through the alternative methods work group is in vitro microphysiological systems. This program was started in the Office of the Chief Scientist 11 years ago with DARPA

to develop these in vitro organ models, and also eventually hopefully a body on a chip.

So this has been a program that we've been involved in from the very first with DARPA, then NCATS joined that program, and I think we have a lot of -- we have some of these chips that are in some of our centers right now, we're doing research on it. We have a user group which Donna chairs where everyone who is doing research on chips across the agency on what they're finding, what their difficulties are, what they might think would be for performance criteria to show if that's working for that particular organ.

And then she has a seminar series, where we bring in outside developers, and they present their chips. So really that user group keeps everybody at the agency informed, aware, and communicating on this one particular technology, and this is sort of -- by trying it on in vitro physiological systems, this is really our first test case to see how effective we can be with this.

And we are also working with the IQ MPS consortium, which is the pharm/tox people from all the centers, and all the companies that are interested in MPS. We're working with them to talk about where we are with each of these technologies, and we're working on ways to talk to our stakeholders about that, and you can see the

first thing that we did was develop a definition for microphysiological systems and organs on a chip, and we're asking all of our stakeholders to write comments on what this definition is, and we have a website where you can communicate with FDA, actually on any issue on alternatives, at alternatives@fda.hhs.gov, and this will be on our website, too, which you saw earlier.

This is a way for this technology and eventually for any alternative technology, you'll know that if FDA is talking about in vitro microphysiological systems, that's almost a tongue twister, or organs on a chip, this is what we mean, because for any new technology, often there's very different names for the same technology. And we wanted to make it very clear that this is what FDA was going to be talking about.

Another thing that we did, we noticed that there was no real meeting place for food chemical toxicology, so we developed a public-private partnership in partner with the Institute for Food Safety and Health at the University of Illinois. They've agreed to be the convener for our food chemical toxicology PPP. And the convener actually is Dr. Brackett, who used to be the center director. This is a partnership with the food industry. It's not pay for play. Anyone can join. So we have food industry, academics, NGOs, we have members from CFSAN, senior members

usually, on the PPP, including members from our research lab.

What we might do, first of all, is look at -- I talked a little about constituents. Constituents are what we call heat-processed contaminants, such as acrylamide, furan, propyl alcohol. There's many different ones. And we can't do animal studies on all of those, but we would like to develop an alternative, sort of a regulatory IATA, to look at these types of chemicals, so we would want to make a generic IATA, or integrated discussant, integrated approach type testing assessment. This, the food industry to just try out and see if this is an alternative approach to looking at a very big problem in food. Things that people have heard about. You've heard about nitrates in food. You've heard about barbecuing and how bad that could be from it. So we're trying to look at using alternative methods and new predictive models to look at that. So hopefully stay tuned and we'll be able to fill you in on that as we move forward with this idea.

Globally, we are also working together on what's called IMARA(?), which is the Global Harmonization of Risk Assessments under EFSA. They put together 40 or 50 food regulators from around the world, and we meet quarterly and try to look at areas where we can harmonize, looking at gaps where we could all work together, and facilitate the

global harmonization of methodology. So CFSAN has put in -- we've talked to them about what we're doing with our predictive tox roadmap, what we're doing with organs on a chip.

We wanted, we brought in another topic that we're asking for discussion on, changing hazard-based risk assessment to exposure-based risk assessment. So you look at the exposure first, and then use that to develop whatever type of hazard criteria or hazard test you need to decide upon the risk of the compound. We also asked them to look at these heat-process constituents that we're finding in food. Let's all work together to look at a method, and then we also brought up or they brought up the topic of micro- and nanoplastics in food. So that's another exciting thing. We're going to help them put together a prioritized agenda so that we can work globally to the food chemistry, food toxicology and work globally on these issues.

What are we doing in our labs? Our lab is the OARSA. I'm blanking on what that stands for. It's the Office of Applied Research and Safety Assessment. We're trying to look at some in silico approaches to prioritize all of these compounds that we have. Like I said, we have cosmetic, we have contaminants, and of course we have botanicals and dietary supplements. One of the things that

they're working on is, is there an in silico QSAR prediction that we can use? And we have the paper to that below. That can give us some ADME information on hepatotoxicity. And then we're also trying to do some in silico modeling to extrapolate in vitro data to in vivo toxic doses. So we're trying to get some way of using our vitro screens that we've developed for several different types of toxicity and give us some idea of what the toxic dose could be and use that as part of a prioritization right now, but eventually, hopefully, a bigger part of our risk assessment.

And the same thing for looking at biomolecular target binding for chemical safety screening. We're trying to identify chemicals in food and dietary supplements with unknown safety and predict the metabolites using in silico techniques, and then, again, another way of trying to identify and prioritize these chemicals for higher testing for maybe more sophisticated in vitro testing, 2D, 3D, chips, or even some targeted animal testing.

Then we also have a chip program at CFSAN. We were the first center to make an agreement with Emulate, which is the spinoff company from the Wyss, the Wyss split, when the DARPA came in to do organs and humans on a chip, they gave two big grants to either the Wyss at Harvard or MIT, and part of that, part of getting that large grant,

about $35 million, was that you had to have a plan for commercializing your chips. They didn't want to give money to academic institutes that then kept that technology for themselves.

So one of the commercialization plans was called Emulate, which is this spinoff from the Wyss at Harvard, and because we had been so intimately involved in this whole thing, Emulate decided that they would put in our lab one of their chips, their liver chip to help beta test that chip before it became a commercialized product.

One of the things we did there, we've been working on it for the last three years and just finishing up, we assessed the system performance, we assessed its ability to actively reproducibly predict hepatic responses, we tried to look at standard endpoints of toxicity, we developed some SOPs that sort of followed GLPs for how you would conduct yourself in the lab, looked at how many -- we looked at some concordance data, we put through hepatic toxicants that we had animal data on and wanted to look at the concordance with that.

And we're just finishing up that research, actually, excitingly, again, Donna is in charge of a bigger CRADA with the same company but one which goes to other ones of our centers that will also contribute to the body

of knowledge that we want. So you see, Donna works at more than just NCTR. We really make her do double duty.

Next, one of the things we're also trying to do is predict ethnic-specific toxicities using hepatotoxic-like cells that we've gotten from iPS cells, induced pluripotent stem cells. Maybe some of you don't remember that several years ago, I was trying to think of the name of the compound, there was a dietary supplement that was safe for everyone except for people of Hawaiian descent. I think OxyELITE was the name of it. It caused severe liver toxicity and even death.

So we realized that was there a way for some of these especially dietary supplements, botanicals, is there a way that we could look at some in vitro ethnic differences and try to predict some of these adverse events? Because remember, we don't have any preapproval authority over these compounds. So that's what we're trying to do with this particular study, establish a panel of human induced pluripotent cells that we can test on multiple ethnicities. We're starting to get a little panel on that, and then we're hoping that we can use this little panel of HLCs in toxicity prediction and testing of CFSAN products. I think we're still developing the panel. We haven't gotten to testing it yet to see whether it's really

an effective tool for us to make some predictions. But we have our fingers crossed.

I mentioned before that we were really interested in worms. I mean, not in worms. We are interested in worms. But in mixtures of metals in children's food. We saw very high levels of arsenic, we also know there's lead, cadmium, and mercury in it. We saw that through our total diet study. We wanted to know if there's some way of looking at the mixtures of those chemicals. All of them are development neurotoxins. We don't really know if they all act, and they probably don't, through the same mechanism of action, whether there's some way of looking at mixtures.

We have a really wonderful study at NCTR that Sherry might talk about where we're looking at the effects of arsenic on develop -- in pups and cognitive ability, and this was sort of a partner to it, can we develop a worm study, a C. elegans study, to look at this and sort of predict -- because even though Sherry's done a wonderful job, it takes a long time to do one of these studies, and this would give us a more effective tool. We knew that some of -- for arsenic, we knew that some of the mechanisms of action and some of the way that arsenic acts, those pathways were conserved in the C. elegans worm, so we thought that this might be a good way to look at the tool.

What we asked them to do was look at arsenic, lead, and mercury alone, but also in combinations with each other, and they saw a developmental delays and they saw changes, developmental delays and changes in motor activity, the worms are running around a lot more, with arsenic alone, and then they're looking at arsenic in combination with these other compounds. So we were interested in inorganic arsenic versus DMA because we had gotten some hints -- DMA is organic metabolite of inorganic arsenic -- we had gotten some suggestions from some of the great studies at NCTR that DMA might be as toxic as inorganic, or even partially as toxic. Understand that when we regulate inorganic arsenic, we have made the assumption that the organic metabolites were not contributing to the toxicity at all, and we didn't put them in our risk assessment, and we wanted to start looking at whether we should.

This is something we're doing under the perinatal thing to look at the, look at one of the short-term PFASs. Also, we're going to be looking at CBD at NCTR in male reproductive toxicity. And finally, I'll just end with we're doing cosmetic -- all our cosmetic research is done at NCTR, and I hope you hear about what we're doing on tattoo inks. This is really exciting. Tattoo inks are cosmetics, but they are not formulated to be used in

humans, they are formulated to be used on texts and stuff, but they are used in humans. NCTR is finding 35 percent were contaminated with microorganisms, and now really excitingly, we want to see if there's transplacental transfer of tattoo pigments across the placenta to see if people that get tattoos or have tattoos, we know some of it becomes systemic, and is that crossing to the placenta and also into the brain.

So that's it and thank you. Sorry I ran a little bit overtime, but we're doing a lot of exciting things, and we hope to use some of these --

DR. ASCHNER: Thank you for the presentation. I don't see any questions from anyone. I'll ask another question then. You talked about the cosmetic issue, it was actually the last slide, about the interaction with the NCTR. But in terms of the liver on the chip, the liver toxicity modeling, certainly the C. elegans, there seem to be a lot more opportunities to collaborate between CFSAN and the NCTR, so I was wondering if you could just elaborate on perhaps a couple other projects.

DR. FITZPATRICK: All of our cosmetic research is being done at NCTR right now. You can see 1,4-dioxane is the contaminant cosmetic, it's a toxic chemical, and we're looking at whether it's a dermal carcinogen, because it's

an oral and inhalation carcinogen. We've been asked to remove that from compounds.

We've got an exciting project that we want to look at bio-printed skin at NCTR, to see if we can have a good in vitro model for skin absorption, model for cosmetics. So I think it's actually a partnership between our cosmetic group and NCTR. We're not doing the cosmetic research in our lab. We're doing all of our research on cosmetics in NCTR, as well as a lot of our arsenic research.

The other exciting thing with Sherry's group too is we're trying to get to build a perinatal program to look at zebrafish and arsenic and cognitive ability, so we can look at the difference between using C. elegans and zebrafish, and then a rodent study, and how we might compare the endpoints for all of those.

Does that answer it? NCTR does a wonderful job on everything, so it's hard not to want to use them for everything that we want to do.

DR. ASCHNER: Yes, thank you very much. Thank you, Suzanne. Are there any questions from Scientific Advisory Board members?

(No response.)

Okay, in this case, we're going to move onto the next presentation. The next speaker is Dana van Bemmel. This is an update from the FDA Center for Tobacco Products.

Please go ahead. You have 20 minutes. We've been pretty good with time, so if you run over a couple minutes, I'll let you go, but if you go more than that, I'll wave my finger, I guess.

DR. VAN BEMMEL: Thank you. Good afternoon, everyone. As Miki said, my name is Dana van Bemmel. I'm the chief of the Research Operations and Advisory Resources branch in the Office of Science in the Center for Tobacco Products, here at FDA.

I'm happy to be here today. I'm excited to take the opportunity to give you a little bit of an update on what our center has been doing, from both our regulatory activities and research activities and hopefully be able to tie those activities to our collaborations and research that we're funding in the portfolio, across the portfolio, but more specifically for this meeting, with NCTR.

The Center for Tobacco Products is unique in that we regulate tobacco products. For those of you who may be newer to this group, that's the regulation of the manufacturing, marketing, and distribution of tobacco products. The overall goal of the Center for Tobacco Products is to reduce the harm from tobacco products across

the entire population. So that includes reducing the number of people who start using tobacco products, encouraging more people to stop using tobacco products, and reducing the adverse health impacts for those who continue to use tobacco products.

Recognizing that not everyone has been on the board for many years, let me just be clear that tobacco products include a number of different products, including products you're traditionally familiar with, combustible direct products -- cigarettes, cigars, and different types, pipe tobacco -- and also includes some of the more popular tobacco products that you've probably seen in the popular press, including electronic nicotine delivery devices, or those devices that are commonly termed as vaping products.

CTP is slightly different than some of the other centers, not just in the products that we regulate, but also in the way that we approach that regulation. We can't take the standard safety and efficacy approach to reviewing and authorizing tobacco products, because we know that tobacco products are inherently dangerous and not safe. So we have this pursuit of a public health standard as we regulate tobacco products. So it's really taking into account the benefits and the risks to both the users and the nonusers of tobacco products, and trying to assess that

overall net population-level impact of the use of tobacco products.

In order to do this, we are, like other centers here at FDA, taking in information, research, and data from all different types of stakeholders. When you think about the landscape of tobacco products and regulation, it's not a new landscape, we're a relatively new center established in 2009, but the research and the tobacco control programs as a whole have been around for many decades. Some of our stakeholders are here on this slide, includes other government regulation, tobacco research at both academic and private institutions, nonprofits, tobacco control programs, and the tobacco industry itself. So all of these represent stakeholders that we engage, and we look to for information, research, to inform our regulatory activities and decision-making.

Like all the centers here at FDA, CTP really has science leading the way in all of our regulatory activities, and in the next few slides, I just want to take the opportunity to talk about some of our regulatory activities. Before I jump to that I'll just note that there are a number of stakeholders as were represented in the other, the previous slide, and those stakeholders are hopefully bringing together the information and the research and data that we need to continue to build that

body of literature to really build what we call tobacco regulatory science. So it's a body of research and information that informs FDA's regulatory authorities around tobacco products.

So to jump into some of those regulatory activities that I mentioned, I'll start with regulation. Some of the more recent examples of regulation related activities that CTP has been involved with include the advance notice of proposed rulemakings, or ANPRMs. Two of the more recent included ANPRMs around flavors in tobacco products and around product standards related to nicotine and tobacco products. So we communicated with our stakeholders, asking and soliciting additional data, as well as looking at the available data that was there, all of this coming together to inform future actions.

The final rule around cigarette health warnings that was recently published as well included a number of different research activities and opportunities that informed that decision-making, including qualitative and quantitative studies.

Another example of regulatory activities at CTP include guidances. Some of the more recent are here listed on the slide. HPHCs are hazardous or potentially hazardous constituents in tobacco. That testing and reporting information including an abbreviated list by product

category on what those constituents in tobacco are, as identified by the literature and the available science.

The ENDS PMTAs, or premarket tobacco application guidance that was recently communicated, contained information on the evidence regarding public health concerns that's needed to -- that is needed and helpful in having included in these premarket applications, and I'll talk a little more about specific enforcement here as our next example, which are the enforcement policies around flavored cartridges and ENDS products.

So the PMTAs, I just want to say, I'm going to get to in a couple of slides when I talk about our pathways for review, but just note that there were guidances or information -- that's really what guidances are -- information to our stakeholders around these various topic areas.

I do want to dive just a little deeper into the enforcement policy around ENDS use. It got a lot of -- it was in the popular press, and I think it's something that we're all pretty familiar with as far as the ENDS use in vaping, epidemic in the United States, particularly among youth.

So in 2019, the end of 2019, FDA and CDC published a study using the National Youth Tobacco Survey results from 2019, and the data from the study really

showed that more than 5 million U.S. middle and high school students were currently using e-cigarettes, having used within the last 30 days, with the majority of those reporting use reporting cartridge products as their usual brand. In addition, the study showed that current e-cigarette users in 2019, of those, approximately 1.6 million were using the product frequently, used on 20 days or more in a 30-day period, with nearly 1 million using e-cigarettes daily.

So with this information, combined with other data from surveys and other current literature on ENDS use in youth, the FDA prioritized the enforcement of ENDS products, specifically those in cartridge-based products. This was on January 2, we issued this policy prioritizing the enforcement against certain unauthorized flavored e-cigarette products that appeal to kids, including fruit and mint flavors. Under this policy, companies that do not cease manufacturer distribution and sale of these unauthorized flavored cartridge-based e-cigarettes, other than tobacco or menthol flavors, risk FDA enforcement.

It took effect in February 2020, and as I noted, it included any flavored cartridge-based ENDS product, excluding tobacco or menthol flavor. Any ENDS -- although this guidance, this policy, enforcement policy, was put forward as a way to address the youth use of ENDS products

and it was very specific to specific flavors and type of product, cartridge-based, the FDA has been very clear in sharing and indicating that any ENDS product that targets to minors or likely to promote use of ENDS use by minors will be looked at by the FDA. So although this was specific policy around those, it does not exclude enforcements if that data should come forward to the FDA that there are other products being used or targeted to minors.

So that was just one example where the data that we are working with, some of which we are publishing with our federal collaborators, as I mentioned the CDC, and other data from other peer-reviewed external sources, really informed an immediate action that CTP took. Some of the other regulatory activities that we perform at the Center for Tobacco Products include tobacco product application review. I'm not going to step through each one of these, but I will define the acronyms for you, because in true government goodness, we have lots of acronyms.

So SE refers to the substantial equivalence pathway. PMTAs, as I mentioned earlier, are the premarket tobacco applications, and the MRTPA stands for modified-risk tobacco product applications. I don't have time to go through what all those pathways are, but if you're interested, there's more information on our website.

I do just want to quickly note two actions that we took around PMTAs in December 2019, so just after this group met last year, the FDA authorized marketing of two new tobacco products through the PMTA pathway, manufactured by 22nd Century, and those are the moonlight tobacco products there in the center of your slide. In addition, in the spring of 2019, FDA also authorized the marketing of a heated tobacco product known as IQOS. It's an electronic device that heats tobacco-filled sticks wrapped with paper to generate nicotine-containing aerosols. So these were two new products to hit the market to be authorized to be sold on the U.S. market through these CTP pathways.

Finally, within the Center for Tobacco Products, we are focused on communication and education and compliance activities. I will note that with the communication activities, it might be difficult to make the connection between work at NCTR and toxicological data and how that informs communication campaigns, but if anyone has seen any of our education campaigns through The Real Cost campaign, some of them are very graphic and they really do target communicating the risks related to specific exposures with using tobacco, including ENDS and cigarettes.

So we worked collectively across the Office of Science and the center with behavioral scientists,

chemists, toxicologists, to really inform the final products that were released with these Real Cost campaigns. The Real Cost campaign leverages a robust media strategy to effectively reach teens and to change their tobacco-related knowledge, attitudes, behaviors and beliefs. So the campaign continues to run nationally.

It airs on TV, radio, print, web, social media, and other advertising avenues. If you happen to be like myself and you are the parent of teenagers, it's not uncommon for me to walk through and see one of these advertisements running through their scroll on their phone through whatever social media they happen to be looking at at the time.

So before I get into some specific examples of NCTR research that we're working on, I did want to just touch on one other activity that really just occurred within the last year, although it feels like a very long time ago when you put it in perspective in the current pandemic that we're working through. But EVALI was just a little over a year ago this time last year. FDA was working with state, federal, and local officials to investigate cases of respiratory illness related to the use of vaping products.

To date, still no one substance has been identified in all of the samples tested, but I think most

of us here at the table anyway and with the board are probably familiar with THC-containing e-cigarettes or vaping products being frequently used in these EVALI cases. In addition, there was a strong link to vitamin E acetate in the EVALI outbreak. So although we have a number of different activities that are happening related to other research activities across CTP and the center, the agency, the vaping and EVALI research does continue. We issued a request for information that recently closed and we're working through that information now. We're also continuing to fund research in this area of vaping related injuries, and so hopefully in meetings to come we'll have some additional data to share on that.

So now in my last five minutes or so, I just want to take a little bit of time to talk to you about some of the past and current NCTR research that we are collaborating with folks down at Little Rock or Jefferson. Most of the research that we are active in right now falls under one of the eight research priority areas that CTP has identified, specifically toxicity. We do overlap in some other research priority areas, but for today's purposes and the currently active research, I am just focusing on the toxicity research interests.

I won't read it to you, but it really is focused on the understanding of tobacco products, their

characteristics, and the toxicities and harm that may be related to those product characteristics. A few of the more recently completed research projects include a bioinformatics project, an in vivo lung toxicity project, and a 14-day nose-only inhalation study of NNK in rodents.

One of our currently active research projects with our collaborators here at CTP includes the pharmacokinetic analysis of nicotine in rats. The study aims are really to establish the pharmacokinetic data on the distribution of nicotine following exposure with three modes of administration. So looking at intravenous oral and inhalation. The benefit to this kind of project, and I did hear that comment early on in the session that it's sometimes difficult to relate the research that we're doing to the regulatory activities. So hopefully I have been touching on some of those here in this talk, but here specifically for this project, it's really to provide data on dose response of nicotines for respiratory and other target organ toxicity, and that information informs tobacco product applications as we review them when they come in. So that would include pathways, all three pathways, that I noted earlier, SE, PMTA, and MRTPA.

Another active project that we have here at NCTR with CTP is the development of a multi-pathway physiological-based pharmacokinetic or PBPK model for

nicotine in humans. This is really a computational tool for nicotine in humans, looking at different databases, literature, unpublished kinetic data, and other resources. The outcome of this work will first be integrated into a computational tool to be developed for nicotine, and then the development of products. It will inform ultimately the development of product standards, specifically nicotine standards and others, as we look to develop product standards that are appropriate for the protection of public health.

A few other active collaborations include an aerosol inhalation project and an early phase PK study. So as you can see, we have been working closely with our collaborators, although some of these may look familiar to some of you, many of them have been ongoing since the last board meeting and before, and like many of the other centers, much of this research couldn't help but be impacted in some ways by the delays that we have with COVID. But we're working to get back in the lab and keep those with our collaborators and keep those moving.

Some potential areas for future CTP collaboration include inhalation tox studies, and that probably may seem a little obvious to this group, given we're tobacco products and the mode of exposure is generally inhalation. So looking at the evaluation of toxicity with repeated

nicotine inhalation exposures might be one project, as well as looking at acute nicotine exposures and adverse outcomes related to those exposures.

Another project that we're currently working through the concept phase with NCTR includes aerosol or nicotine delivery systems, and so aerosol exposure with an ALI model, and I'm really excited about this project. I think it has a lot of potential to inform a number of review and regulatory, other regulatory activities that we do here at CTP, but having the ability to look at cytotoxicity, genotoxicity, and simulations of human inhalation exposures with this type of ALI, culture ALI model, could really be beneficial to the tobacco regulatory science field.

Finally, we are currently -- another potential area of collaboration could be around flavors in tobacco products, and actually Suzy did a nice job of introducing this idea that flavors come -- the chemicals that comprise flavors come with some of their own harms and toxicities. So having the ability to look at those in various toxicity models is very valuable as we look through the premarket applications that are coming to us from the tobacco industry.

So I think my time is up. I just want to end by thanking our key collaborators. I didn't -- I listed each

of the individual collaborators by project on the slide, but Brad, Krysti, Jonathan, and Keyur are really instrumental in making sure that all of these projects stay on track and keep moving forward.

Then I will leave you with this slide for your reference. I'm happy to take any questions.

DR. ASCHNER: Thank you, Dana. Thank you very much. We have one question. I'll read it to you. COVID-19 impacts the convalescent cardiac and pulmonary disease, even in convalescent patients who may not have had significant disease. Is CTP considering the potential of exaggerated injury and response to nicotine or tobacco products in general? Influenza has some of this potential, but COVID-19 may be worse and perhaps the risk may be more persistent. So I guess basically the question is are you considering the potential of exaggerated injury and response to nicotine in these patients?

DR. VAN BEMMEL: Yes, it is a good question. I can't speak specifically to all of the different activities that our individual health science division is working through with respect to that, but I can say that within our research portfolio, in addition to our work with NCTR, we have a collaboration with the NIH to fund a large grants portfolio, and I know that there are individuals that are

looking at that kind of impact, potential impact of nicotine and COVID.

So I don't have any information to share on that, but I have heard of that, I am aware of it, and I know that there are folks within the research community who are looking at that, even if it's not being directed directly by FDA. Some of these things are just moving really quickly, and quite frankly the academic institutions have flexibility to move on some of these things, to leverage some of the resources that they have a little more quickly.

DR. ASCHNER: Thank you.

Greg, if you want to follow up, why don't you just unmute yourself or if you have a comment. I think it's going to be a lot more interactive than waiting for people to type.

DR. LANZA: The reason is that I worry a lot of the patients that we're seeing, particularly using MRI to characterize, but we're seeing patients who are barely mildly symptomatic, maybe not even hospitalized, having secondary problems in their heart and certainly in the lungs is possible. So I'm worried about these younger people who are using these products in increasing numbers according to your presentation, actually being at far more risk than they realize and that we realize, and then when they get the second hit, whatever it might be that's

prothrombotic or myocarditic or interstitial lung disease or whatever, now their response is worse and our ability to treat it jumps from being something mild to something much more severe. It's just an unknown territory, but there are so many people involved in this case in the pandemic. That's why I was asking the question.

DR. VAN BEMMEL: Yeah, again, it is a great comment, a great question. I made some notes here that I'll take back with me. I will say that it's been several months now, but FDA did work with our federal partners at NIH and CDC to pull together some language to share with the public around the use of tobacco products and the potential risks associated with COVID, specifically COVID being diagnosed, and the additional risks severity of the disease, but you're right, it's uncharted territory.

So really, I think the message is that using tobacco products is never safe and then in light of the pandemic, there may be additional risks. So your point is well taken, and I will take that back with me.

DR. ASCHNER: Thank you. I would like to make a suggestion, actually, as we move on. If you have a question, why don't you just type questions, I'll see your name, and I'll give you the floor. I think it's going to be a lot more interactive, you can follow up, and it will

generate a lot more discussion. So let's try to practice this new approach if possible.

Any other questions?

(No response.)

Okay, if not, we're scheduled for a break. It was originally 20 minutes, but we are running a few minutes late. So we'll take just a 10-minute break. We had one not too long ago. We'll come back in 10 minutes. Let's make it 10 minutes, Michael.

Great, thank you.

(Break.)

DR. ASCHNER: Okay, the next presentation, well, you can see the title, Dr. Tan. I see you had time to change. Go ahead.

DR. TAN: Good afternoon, everyone. Thank you for the opportunity to speak. I am the director for the Office of Research for the Center for Veterinary Medicine, and I'm going to give you a little bit of an introduction to us. The Center for Veterinary Medicine is responsible for assuring that animal drugs and feeds are safe and effective and that food from treated animals is safe to eat. This authority is derived from the Federal Food, Drug, and Cosmetic Act.

The Act was amended in 1968 to include sections which specifically address animal drugs. These amendments

were designed to ensure that animal drugs are safe and effective for their intended uses and that they do not result in unsafe residues in foods. The mission statement for CVM reads protecting human and animal health and to achieve this mission, we make sure an animal drug is safe and effective before approving it. The center approves animal drugs for companion animals such as dogs, cats, and horses, and for food producing animals such as cattle, pigs, chickens, and honeybees. If the drug is for a food-producing animal, before approving it the center also makes sure that food products made from treated animals like meat, milk, eggs, and honey are safe for people to eat.

We monitor the safety and effectiveness of animal drugs in the market. We make sure animal food, which includes animal feed, pet food, and pet treats is safe, made under sanitary conditions, and properly labeled. We make sure food additive use in animal food is safe and effective before approving it. We help make more animal drugs legally available for minor species, such as fish, hamsters, and parrots, and for minor uses in major species, such as cattle, turkeys, and dogs. We conduct research that helps the center ensure the safety in animal drugs, animal food, and food products made from animals.

At CVM, we're very lucky to have the state-of-the-art research complex with offices, laboratories, animal

buildings, and pastures. This facility includes mass spectrometry, microbiology, whole genome sequencing, and stem cell laboratories, analytical instrument rooms, a radiolabeled materials lab, and many specialized laboratories designed for multidisciplinary studies.

The animal research buildings accommodate beef cattle, dairy cattle, calves, swine, sheep, poultry, and a variety of aquatic species. Our investigators at the Office of Research possess expertise and a wide variety of scientific disciplines, including veterinary medicine, animal science, biology, stem cell physiology, molecular biology, chemistry genomics, proteomics, microbiology, immunology, physiology, epidemiology, pathology, aquaculture, and pharmacology.

I'll run over at a high level some of our collaborations with NCTR. You'll also see some of our staff. We have found recently that as a laboratory community we're very much used to being together, and with our current situation, we are not quite as together as we normally are, and we find pictures of one another really does help. So I'm sharing this with you today.

Okay, advance safety assessments of FDA-regulated products using high-throughput and high-content quantitative approaches to cultured human cells to evaluate genotoxicity. There are a lot of collaborators on this,

notably Xiaoqing Guo from NCTR, Tong Zhou from CVM, and from CDER, Timothy Robison. NCTR conducts in vitro toxicology assays consistent with standards of the National Toxicology Program, and has experience in working with nanoparticulates. For us, the impact of the work is that in vitro genotoxicity studies are routinely used as screens contributing to the evaluation of the potential carcinogenicity of new animal drugs. As nanoparticulates find their way into new animal drug production, it's important to know their impact on these assays.

Evaluation of virulence potential of bacterial pathogens using three-dimensional tissue culture model systems. Again, we have a lot of collaborators, Bijay Khajanchi, Heather Harbottle from CVM, and Chris Whitehouse, who is also the new deputy director for the Office of Research at the Center for Veterinary Medicine.

NCTR developed a proposal to establish a 3D tissue culture model to evaluate the virulence of bacterial pathogens, particularly salmonella, utilizing their expertise in cell culture systems of bacterial pathogens and bacterial virulence. The impact of this work for us is to help us better evaluate premarket microbial food safety risk assessments. It supports effectiveness studies of nontraditional drugs as alternative strategies to combat microbial pathogens, evaluate antivirulence alternatives to

antibiotics, and evaluate virulence properties of bacteria associated as part of the national antimicrobial resistance program.

Evaluation of cadmium oxide nanoparticles as a nanoparticle type positive control for in vitro toxicity assays. We have as our collaborators Tao Chen from NCTR and Tong Zhou from CVM. NCTR conducts in vitro toxicology assays consistent with standards of the National Toxicology Program and has experience in working with nanoparticulates.

The impact for us is in vitro genotoxicity studies are routinely used as a screen contributing to the evaluation of the potential carcinogenicity of new animal drugs. As nanoparticulates find their way into new animal drug production, it's important to know their impact on these assays.

Salmonella enterica virulence and plasmid characterization databases and analysis tool development. NCTR is aiding in the development of a national virulence gene database, and this database will be used by NCBI to characterize bacteria based on genomic sequence data and help refine hazard characterization for microbiological risk assessment.

Studies on the intrinsic structural multidrug efflux pump mechanisms in antimicrobial resistance,

salmonella enterica, and their role in antimicrobial resistance. NCTR has expertise in bacterial efflux pumps and will conduct the laboratory work to evaluate their contribution to antimicrobial resistance in salmonella enterica, and for us, the work will enhance genomic-based AMR surveillance by helping to ensure a complete AMR gene database is curated. For this, we thank Ashraf Kahn, Mohamed Nawaz, Steve Foley, and from CVM, Shaohua Zhao.

Evaluation of virulence potential of bacterial pathogens using three-dimensional tissue culture model systems. This 3D culture model system will more closely mimic in vivo conditions than conventional 2D cell culture. The system will more accurately assess the virulence in important bacterial pathogens and evaluate potential new therapeutics that target virulence mechanisms of different pathogens. Efficient models to assess the virulence of pathogens could facilitate studies that provide a better understanding of the mechanisms of action of novel anti-virulence drugs. Proposed research will help FDA determine the three types of data, the types of data, needed to appropriately evaluate these new classes of drugs. Our collaborators are Bijay Khajanchi, Heather Harbottle, Jeff Gilbert, and Chris Whitehouse.

This is a high-level overview. We do have a lot of demonstrated success in collaborations. You'll see here

there's quite a few. I'm not expecting you to read these. Some of the conversations that I've heard back and forth today are how would we continue to enhance collaboration with NCTR? So I am about a year into my role, and what we're doing is we're reevaluating how we approach collaborations. We've heard today both PIs get together and innovate together. We've also heard top down. I think what we're going to be looking for at CVM is a hybrid of both.

We'd like to encourage the PIs to work together. There's a lot of richness there when the PIs get together and bring something to that. But at the same time, we would also like to have management involvement, and we very much welcome coordination with NCTR so that we can make sure that we're connected in with the work that our PIs are generating together and the ideas that our PIs are generating together.

With that, I'm happy to take questions.

DR. ASCHNER: Thank you very much. Are there any questions from Scientific Advisory Board?

(No response.)

Hearing none, I guess we'll proceed. Thank you, Dr. Tan.

DR. TAN: Thank you.

DR. ASCHNER: Thank you very much for a concise presentation. Our next speaker is Dr. McMeekin, Judy McMeekin, and this is coming to us from ORA, and I had to learn all these acronyms as well. So please go ahead.

DR. MCMEEKIN: Thank you very much for inviting me to be with you. It is my honor to be here representing FDA as associate commissioner for regulatory affairs, a position that I assumed just this past April. So today, I will provide you with an update on the Office of Regulatory Affairs, or ORA, our priorities, and our accomplishments, and outline how our scientific research efforts intersect with those of NCTR and discuss our plans for the future.

But first I would like to give a quick overview of ORA for those of you who may not be as familiar with our organization. So the Office of Regulatory Affairs, or ORA, advances the FDA's mission by conducting field operational activities on the FDA regulated products to ensure their safety, effectiveness, and quality. As FDA's lead office for all agency regulatory field activities, ORA is responsible for a wide range of mission critical activities, including inspections and investigations, including criminal investigations, and also sample collections and analyses, along with examination of the FDA regulated products that are offered for import into the United States, oversight of recalls and execution of

enforcement actions, response to consumer complaints and emergencies, and also development and promotion of state and local partnerships as they work with us on inspections.

The FDA regulated products account for 20 cents of every dollar spent in the United States, and ORA protects consumers and enhances public health by maximizing compliance and minimizing risk of all FDA regulated products. This includes human and animal foods, cosmetics, dietary supplements, human and veterinary drugs, vaccines, blood products, tissue, tissue products, allergenics, cellular and gene therapy products, along with medical devices and products that emit radiation, followed up by tobacco products.

ORA has staff in 231 offices across 49 states, including the Commonwealth of Puerto Rico, with staff both temporarily and permanently assigned to foreign posts also. ORA manages 13 scientific labs, all overseen by Dr. Paul Norris, who is the director of ORA's Office of Regulatory Science, or ORS. ORA also develops and maintains information technology systems that are used across FDA that promote efficiency through information sharing and enable operational processes and decision-making by employing risk-based tools.

In addition, ORA promotes an integrated food safety system, or IFSS, by providing resources to our

state, local, and tribal and territorial regulatory jurisdictions to conduct inspections, collect samples, share information, and enhance program capacity and infrastructure. So we rely very heavily on our state and local regulatory colleagues.

ORA's research arm is the Office of Regulatory Science, or ORS. As I mentioned earlier, ORS has 16 laboratory programs at 13 different locations across the nation. Major areas of research include food, feed, pharmaceuticals, cosmetics, medical products, tobacco, and other specialty labs. The labs perform highly specialized analyses of domestic and imported products. Several also conduct research to advance regulatory science.

It is important to ensure that ORA's research aligns with our mission, prioritizes high impact projects, and has measurable outcomes. Within ORS, the Office of Research Coordination, Evaluation, and Training, or ORCET, they manage the research conducted in the ORS labs. Dr. Selen Stromgren oversees the Office of Research Coordination, Evaluation, and Training. In order to do this, this group works with FDA centers and other stakeholders to identify priorities and directions, evaluate proposed research for technical merit, mission relevance, and deliverable results, and quantitatively track and assess outcomes.

So ORA's primary scientific mission is to support agency preventative and enforcement action through our laboratory findings. The ORA's science strategic plan reflects long-term tactical goals to uphold that vision. It prioritizes the quality and integrity of science with a focus on defensible results and methods that are investigative in nature. It also calls for laboratory capacity building for maximum efficiency, which we can accomplish by partnering with our states.

The strategic plan also prioritizes horizon scanning for project needs and developing tools that allow us to respond rapidly and nimbly to emergencies. It is not always easy to predict what will hit us next. This COVID-19 pandemic has brought into the spotlight issues such as operational readiness and development of functionality and quality tests on urgently needed PPEs, opening the door for marketing of products by unregistered firms under the emergency use authorization umbrella.

COVID-19 has undoubtedly required us to shift our focus and to reprioritize our work so that we can be most helpful to response efforts. For example, research on testing for attributes of advanced pharmaceutical products and novel microbial agents, such as various foodborne viruses, but despite the change in focus, we also have made

considerable progress towards our ongoing priorities, including scientific research.

In alignment with the ORA science strategic plan, we continue to prioritize high risk food examination and sampling at the ports of entry. This has included findings of microbiological pathogens of salmonella and listeria monocytogenes in several imported food products. We have also created or adjusted import alerts and flags on our PREDICT model, which is a risk-based import screening tool, so that foreign manufacturers' products can be sampled at ports of entry. For instance, we are currently working on a large hand sanitizer assignment from CDER. We are testing imported sanitizers for label compliance of ingredients and have found violations, such as less than the indicated ethanol and the presence of methanol. We also have the capability to feed these lab findings back into PREDICT to impact risk scores of the companies sending their relative products.

The COVID-19 pandemic has also not deterred us from investing in our infrastructure. Recently, we kicked off a multiyear effort to upgrade our labs, opening a new facility in Alameda, California for our district and laboratory workforce. We also have construction of the new Kansas City laboratory that will be completed in September of this year. This new facility will have much larger

cubicle spaces than the standard size, which will significantly improve our ability to follow CDC guidelines for social distancing.

We are also exploring touchless options for doors and restroom faucets, as well as the incorporation of UV lights that kill bacteria, molds, and viruses. The new Winchester Engineering and Analytical Center, many of you may know that as WEAC, is currently under construction and it's scheduled to be completed in 2021. We are also relocating the Atlanta laboratory and district office, as well as expanding our forensic chemistry center that is located in Cincinnati, and that should be completed in 2022.

This pandemic has presented opportunities for us also, opportunities to innovate and reassess how we conduct our work, and as the products we are asked to review become more complex and specialized, we see a larger demand to actually develop innovative technologies and methods. As we look to the future, ORA will continue to rely on collaborations with trusted partners like NCTR, whose research strengthens our risk-based decision-making. The unique scientific expertise in NCTR will continue to be key in reinforcing FDA product centers and their regulatory roles.

Many of you are aware that ORA and NCTR have joint membership on several agency-level committees. These include but are not limited to the agency-level nano taskforce, which is chaired by NCTR, the FDA mass spectrometry workgroup, which is chaired by ORA, and the artificial intelligence, or AI, workgroup. This is chaired by NCTR. ORA is involved in multiple collaborations with NCTR using cutting-edge science and technology to address some of the most pressing public health issues of today. Of note, NCTR has a large interest in standing up the artificial intelligence-based applications, or AI-based applications, with research mostly focused on pattern recognition in images. They are looking to ORA for potential application areas such as identification of species of pests in our field program.

NCTR's AI program can identify the species using images of fragments, like wings or legs, which tells us how risky or detrimental the infestation is to the product it was found in. Another application is rapidly reading labels at our international mail facilities, and this would be helpful in telling us if the product that is being mailed in is counterfeit, a similar application to the FCC's counterfeit detector device. This is really encouraging for us and would help increase efficiency at our international mail facilities.

The third application concerns patterns in mass spectrometry data. In our Persistent Pollutants Program, we generate a lot of data, and being able to reliably tell whether certain data are high quality helps us decide what datasets to actually accept and reject.

So I'm excited about areas of potential collaboration with NCTR. Of note, I see an opportunity for continuous partnership on scientific initiatives that include other agencies with a focus on nanoplastics and artificial intelligence. NCTR has helped us this past year on a homeopathic product that contains an ingredient of unknown toxicity. The NCTR scientists conducted a literature search and summarized their findings on the expected toxicity, which helped the collecting district and CDER compliance with their next steps.

So we first see having similar interactions with NCTR in the area of toxicology assessment. So now just a little bit more on ORA's recent activities and future plans. We have been working on a data-driven approach to help us make informed risk-based decisions on resuming our prioritized domestic inspections. These inspections are being prioritized based on the risk of a firm, the product that is being produced, and any other factors developed in concert with our center colleagues.

In addition, when deciding which facilities to inspect, the FDA will consider the health and safety of our staff. Until further notice, we are using the FDA developed COVID-19 FDA advisory system to help us make decisions on resuming inspections in specific locations. The advisory system qualitatively indicates the status of COVID-19 cases in a specific local area based on county, state, and national criteria. Its metrics are based on the phase that the state is in, as defined by the White House guidelines for Opening Up America Again, and information measured at the county level indicating the current trend and intensity of infection. This COVID-19 FDA advisory system, along with other factors like state travel restrictions, help us identify when and where to resume prioritized domestic inspections, conduct investigations, and conduct sample collections and analysis.

We also continue to improve our core operations. We cannot achieve our goal of reducing the occurrence of food-borne illness risk factors without partners such as NCTR. Working together, we are confident that we can bend the food-borne illness curve, but we must innovate and employ different strategies than we have had in the past.

Of note, we are implementing the blueprint for A New Era of Smarter Food Safety. This is an FDA initiative shaped by valuable input from our stakeholder partners.

The new era is charting the course to enable us to keep pace with the many anticipated changes in the food system over the next 10 years. There are four core elements of the new era, which are tech-enabled traceability, smarter tools and approaches for prevention and outbreak response, new business models and retail modernization, and food safety culture. We'll share more details in the coming months on this initiative.

As a strong integrated food safety system is essential to a safe food supply, domestic mutual reliance is also a key component of the new era of smarter food safety initiatives. The essence of domestic mutual reliance is partnerships, which are never more necessary than during challenging times as these. Sample collection and laboratory capacity analysis and recording are critical to achieving true mutual reliance. We are always looking for new and creative ways to engage and invite your feedback on how you think we can reach decisions.

ORA continues to leverage technological advances by exploring new platforms to do our work more efficiently. Among many other ideas, we are looking at how to integrate machine learning capabilities into our processes to identify and implement solutions that make our current processes more efficient.

Innovations like this will support all of our operational components and programs and set us up for success in our everchanging world. So I'd like to thank you again for your partnership and shared commitment to public health. I appreciate all of the collaboration, not only with NCTR but also with our other center laboratories. I'd now like to turn it over to NCTR division directors to provide an overview of their research activities. So thank you very much.

DR. ASCHNER: Thank you very much, Judy. I'd like to open the floor to SAB members. Do you have any questions?

I should mention what we decided to do a couple of years ago actually was to have the presentations from the different centers, the FDA centers, before the individual divisions at NCTR. We thought it would give everybody much better background, especially the SAB. So as we hear the division presentations, we would be able to perhaps find opportunities for these divisions to collaborate to a greater extent with the different centers and so forth. So the format was changed. This was the last one, actually, of the presentations by the different FDA centers. So this is a good opportunity if anybody has any comments about integrating between different centers or any other comments, this is the time to mention it.

DR. LANZA: This is Greg. I just had one general thought over the last several presentations, and I would like to bring it up here. That's basically how much of the collaboration between NCTR and the different centers could be very focused protocols and specific areas in response to either questions that are occurring up in the center or initiated by NCTR, versus some things that are a bigger level of expertise developed at NCTR. An example might be the nanotechnology center, that would serve as a level of expertise for all the centers, and deal with some of these problems, not just what's current, but what's actually more future, because there are a lot of projects that are legacy projects that have continued to develop the science but may not be dealing with the bigger questions and level of expertise needed in the future, and that expertise wouldn't happen in a heartbeat just because you want to start. So how do you feel about this?

DR. MENDRICK: Bill Slikker, this is Donna Mendrick. I think you should answer that question.

DR. SLIKKER: Greg, I'd be glad to provide some words about the question. I mean, you know, we have the opportunity for the sort of essence or the questions to be raised to come from either the product line centers or from the NCTR or in some cases from the advisory group that you're a member of, and all these different areas can

provide interesting questions, and then it's a matter of regarding that particular question being if there's a general FDA reason, you might say, for doing it, if it's within the scope of the FDA's mission, then we could certainly pursue that, and we do that in a very systematic fashion. What we do is that we begin with a concept, which is a two page summary of what we'd like to do, has some background material in it, poses a question, talks about maybe some general resources that will be necessary, and what the real impact would be on FDA, and then if that is reviewed, which it is, not only internally at NCTR, but then goes to other centers that have common interests, and we get their feedback, if that one is then said to be, well, yes, this is really important issue for FDA, then it can be pursued, but sometimes it's not. Sometimes it's like, well, no, we're already doing that or that's not important today. Then it's set aside.

For those that get the greenlight, then a full proposal is generated, and that's also evaluated by those same centers within the FDA, and input is received to improve it, oftentimes we can find collaborators in other centers that want to join in or be part of it and help devise even a better protocol, and at that level, then after approval going through all the different animal care and use or human use, and other kinds of safety

considerations, it will finally be approved and funded for the study to begin.

So that's the process that we use. So at any one of those opportunities there, Greg, as you know, there can be input from the various centers or from the FDA, NCTR staff or from others even outside the agency, to look at certain areas that could be of interest to FDA, and that decisional process of whether or not it moves forward depends on whether or not the question is really of mission relevance to FDA. So does that help some, Greg?

DR. LANZA: It does. But let me give you an example. So we have heard several different centers talk about artificial intelligence in different ways. A lot of it's just really the most basic automation, pattern recognition. But the power is much greater, and particularly with 5G coming up, and I just wonder whether this is a situation where the different centers could keep expertise, could invest some of the funds into NCTR to develop a greater expertise that would be available to all the FDA without -- because the ability of people in that field is very flexible and I just wonder whether you're not duplicating the types of people in different centers that maybe should be at NCTR.

That's just one example, but people who work in that area are not so much subject dependent as they are the

expertise in using the different languages they use in their approach to nodal networks. That's what I'm getting at, that kind of project. That's what you did with the nano project. That worked across all centers, anything that's nano small stuff, whether it was for degradants like the plastics that you're doing now, or drug delivery or contrast agents or anything. I'm just wondering as I listen whether there isn't enough investment by the different centers chipping in to NCTR to build expertise in areas that are being pursued in all the different centers.

DR. SLIKKER: It's a very good point, Greg. We have to understand that FDA is a relatively large sort of multipurpose kind of agency, because you do have rules and regulations for drugs that are different from foods, different for devices, and that all those different rules and regulations are really the focus of the various centers who have that responsibility and mission. So there is some opportunity there for building alliances and collaborations, and as you learned about, we do have the working group on artificial intelligence.

We also have ones on newer technologies, et cetera, as many of these working groups, and those tend to be across the entire agency, with representatives from the various centers and ORA to have input into those working

groups to try to build some of a structure in the approach as you're speaking, sort of the strategy.

But we have to realize that even though we do have a very fine NanoCore facility that's actually built up with different kinds of support from ORA from NCTR and some from even the National Toxicology Program, that there are other activities within other centers as mentioned with CDRH that they also have a nano facility as well, and so these are shared amongst the various centers and we do certainly make those kinds of capabilities available across the entire FDA. So in some cases, you will find things coalescing in one center or another, but oftentimes the application of the technology has to be agile enough to be able to fit into a food issue for foods and a drug issue for drugs and a biological issue for biologics, and therefore there's a certain expertise needed there as well.

So in some cases, FDA comes together to build these particular kinds of approaches that are sort of systematically developed in a strategy way that is sort of brings folks together; in other cases some of that expertise resides in the various centers, which is appropriate as well. So we have a combination of what's achieving that goal.

DR. MCMEEKIN: Bill, this is Judy from ORA, and I think that times, meetings like this are helpful, just so

that in sharing the experiences and knowing where some of the expertise is so that when we do have issues such as utilizing AI, that we can think of like where else are the experts within the agency so that we can bring those on to those working groups to really leverage that expertise.

DR. SLIKKER: You are right, Judy. The idea of sharing information is key to understanding all these possibilities. In some cases, the project is so large that you almost have to think about finding another source of that kind of manpower or that kind of approach. However, what we have found over the years that in terms of our bioinformatics biostatistical division, it has been able to find many partners within FDA where they wanted a very defined software approach using AI and other bioinformatic tools to achieve that, and they have been very successful in meeting those particular goals.

So it depends on the magnitude of the job that's at hand. It depends on how much interaction they really want, how personalized you might say that they want the instrument that's being developed, and all those things influence, so where you would turn for support, but when it comes to partnerships, that's a key part, knowing where the expertise is, being able to mobilize it for a particular purpose, and that's what NCTR wants to support.

DR. ASCHNER: Thank you, Bill. Thank you, Greg, as well for the question and for the good discussion.

We are going to move on. Our next speaker -- we are going to move now to the different divisions. So we'll do, I believe, three of them today and then we'll finish the rest of them tomorrow.

The first division is the Division of Biochemical Toxicology, and we'll hear a presentation by Dr. Fred Beland, who is the director of this division.

Fred, please go ahead.

Agenda Item: NCTR Division Directors: Overview of Research Activities

DR. BELAND: I am going to give an overview of the Division of Biochemical Toxicology. The first few slides will be -- you're going to see a template that Donna Mendrick asked us to use. This gives an overview of how many people are in the division. You can see we have 30 research scientists, fulltime positions. We have nine support scientists. We have two administrative people, and we also have postdoctoral fellows. This is the number totals as 46. It's been pretty constant over the years. We maybe have a few people slightly less than we've had before.

I think the question was asked, I think by Dr. Aschner this morning how are we coping with the coronavirus

situation. Throughout, since about the middle of March, I would say we have about 25 percent of the people within the division will be here each day. Some people will be here five days a week. Others have not -- the people who do molecular modeling or computational modeling, they have not been in NCTR since the start of the pandemic.

We work very carefully. Most people are working individually in laboratories. So we have adequate separation. We take adequate precautions. We follow all the guidelines that have been issued by the FDA headquarters.

As far as collaborations, this division collaborates with each of the other divisions and NCTR. We also collaborate with all of the product centers. So we have CBER, CDER, CDRH, and so forth. We have had a very longstanding interaction with the National Toxicology Program. We get funding from the NCI. We sit on advisory panels for the EPA. We have been involved with CDC.

We also have a lot of global impact. A number of us have been working with IARC in Leone, the World Health Organization, with the European Food Safety Administration, with OECD and the Japanese Food Science Agency.

This gives our mission and goals and strategies. I think the goal is probably the most important is that what we try to do is we try to characterize toxicity and

the carcinogenic hazards of chemicals, especially those that are of interest to the FDA. Traditionally we have done this through sort of a pattern or an approach where we conduct chronic bioassays. We have a long history of doing that, a lot of which have been funded by the National Toxicology Program.

We also do mechanistic studies to try to determine whether or not what we are observing in the bioassay are pertinent to what goes on in humans. Then I would say ten or so years ago, we really stepped up our computational modeling so we could take the animal bioassay data, we could take the mechanistic data, and we could do computational modeling with, again, the idea of extrapolating from animal studies or in vitro studies to what goes on in humans.

I have one slide, just we're asked to talk about the most important accomplishments in the last year, and I would like just to emphasize three things here. The first is for many, many years, we received very substantial funding from the National Toxicology Program to conduct bioassays, mechanistic studies, pharmacokinetic studies on bisphenol-A. Finally at the end of last year, we published an NTP report, but this is also now out in Food and Chemical Toxicology. The lead author is Luisa Camacho and Barry Delclose was also a major, major driver.

So the last piece of this puzzle that needs to be put together is an integrated report, because in addition to the work that was conducted here on a GLP bioassay, there were a lot of mechanistic studies that were conducted by a group of academic researchers. So there's supposed to be an integrated report put together. There's a bit of controversy concerning this. So I'm ready to move on. I think we've furnished CFSAN with the information they needed from the bioassay, and I think it's time to move on to something else.

We've done -- under the direction of Dan Doerge and Nathan Twaddle at analytical chemistry, did most of this work, we've done a lot of pharmacokinetic studies of inorganic arsenic and also organic arsenic in mice, with the intent of using the pharmacokinetic data to help design a chronic bioassay. These pharmacokinetic studies have been published in at least six papers now. There's a few more that -- there's one that has just been accepted, and I think there's one more that is being written.

I think there was the all the pharmacokinetic data were used to construct a bioassay. Unfortunately, we've had trouble securing funding to do the bioassay, and at the present time, I suspect it's not going to be done.

The third area -- this is a new paper that was just submitted for publication in Archives of Toxicology by

Si Chen and her collaborator Lei Guo, and the idea behind this is a lot of toxicity studies are conducted with HepG2 cells, human liver cells, and the problem with HepG2 cells is they really -- they don't have, they have very poor cytochrome P-450 activity. And since metabolism can be intimately involved in liver toxicity, Si Chen and Lei Guo thought it would be very worthwhile to have HepG2 cells that were transfected with cytochrome P-450. So they have put together a panel of stably transfected HepG2 cells that contain -- each contains an individual P-450, and they have a total of 14 cell lines.

This is just a sampling of what we have done. I looked at my midyear review for this year, and so far, division members have published, have 40 papers that have been published with a 2020 publication date.

What I'd really like to deal with for the remainder of the talk is what we're currently doing, and we're currently doing, these are projects that have started for the most part within the last six months, since the first of the year. Some of them are a bit more mature than others, but what I figured is if this a Science Advisory Board and you want to provide advice, these are the projects where we would welcome any advice you have as far as the direction or the things that we're missing that we should be doing.

So I want to talk about three areas. I want to talk about tattoo pigments, I want to talk about CBD or cannabidiol, and then talk about COVID-19.

Suzy Fitzpatrick mentioned the interest that CFSAN has in tattoos or in tattoo pigments, I should say. This sort of outlines the problem, from the way that CFSAN and we are looking at it in collaboration. Tattooing is very prevalent in America, now, with perhaps 50 percent of the population, and a lot of the people who are getting tattoos are women of childbearing age. What's not recognized is that a tattoo can contain 250 milligrams of ink and the average person who is tattooed may have greater than four tattoos. So there is about a gram of material being placed under the skin of these individuals.

What's not recognized is most of the ink or pigment disappears, and it's really not known where it goes. So I'm going to talk about two projects, one that deals with the fact that women of childbearing age get tattoos, they can become pregnant, and the question is where are these tattoo pigments if they're migrating from the tattoo, where are they going?

So this is the first project, and this was done in conjunction with CFSAN. CFSAN had this problem. They wanted for us to help them address it. The funding comes

from the Perinatal Health Center of Excellence that Bill Slikker mentioned earlier today.

What we're doing is, and the principal investigator on this project is Mary Boudreau. What we are doing is we are looking at three pigments, Pigment Orange 13, Pigment Yellow 13, and Pigment Red 22. These pigments will be administered to SKH-1 mice. SKH-1 is a hairless mouse that we have used for phototoxicity studies. We're administering carbon-14-labeled pigment, and then what we are going to do is assess what where the pigments end up, and the way this is done, they'll be tattooed, the mothers will be tattooed on gestational day 1, they'll be carried until gestational day 18, at which time the fetuses will be taken, and then we'll do a complete survey of where the radioactivity has migrated.

Where we stand on this project now is Mary Boudreau and Michelle Vanlandingham are learning how to tattoo. It's not a trivial process in a very small mouse. It is our intent to get about 15 milligrams of ink applied to the animals, and this will contain about 50 microcuries of radioactivity. So I have seen some of the mice that have been tattooed. What you can picture is the whole back of a mouse just with a big orange blanket on it. It's a solid, very solid tattoo.

The second study is a very similar study, except we're not going to be using pregnant animals. The question is what are the long-term persistent effects of these tattoo pigments. So even though there's three listed on this slide, we're going to start with pigment red, and see over a period of 20 weeks, what happens to this tattoo. Where does the pigment migrate? Again, this will be done with radiolabeled material. The idea is again to give the same amount of material we give to the pregnant animals and with the same amount of radioactivity.

This again, I should point out, has been requested by CFSAN and has been funded by CFSAN. Where we stand with this project is we intend to -- assuming we get the tattoo procedures worked out, we intend to start tattooing around the start of the new fiscal year.

Okay, cannabidiol. The cannabidiol projects I'll talk about have really started. We started having discussions with both CFSAN and the Center for Drugs towards the end of the calendar year last year.

MICHAELKAWCZYNSKI: I will pull it back up, sorry. Somebody accidentally ended it. Bear with me. I am just making sure I grab the right one.

(Pause.)

DR. BELAND: The use of CBD was approved in a drug called Epidiolex in 2018 for the treatment of certain forms

of seizures. If you look at the approval, there were questions regarding the pharmacokinetics that CDER still felt should be addressed. So that's one issue. Another issue, if CBD containing material is being added to food, and yet it's not known are there potential toxicities associated with it. In addition, there's been a large number of cosmetic products have CBD as being added to.

So there's two issues here. One is the issue of the oral administration of CBD and a need for federal pharmacokinetic data, and the second issue is there was a need for dermal pharmacokinetic data, which was just nonexistent. Then the last issue is there's some indication that there may be some toxic effects on the male reproductive -- that CBD may cause toxic effects on the male reproductive system.

So based upon this, we have initiated four projects. Again, this was through discussions with both CFSAN and CDER and then a CBD working group. The first thing I'd like to point out is that this slide shows the structure of CBD. It undergoes metabolism on this methyl group at the top of the molecule to go to 7-hydroxy-CBD, and then a subsequent oxidation gives you 7-carboxy-CBD. The important thing about this is in humans 7-carboxy-CBD is the major metabolite. So the rodent data tends to emphasize the 7-hydroxy, but for humans it's the 7-carboxy

that's the major metabolite. So in order to get a good estimation of what's happening, you really need to consider both the hydroxy and the carboxy metabolites.

So the first project that I'd like to talk about, this being directed by Si Chen, and it's an in vitro study to look at male reproductive toxicology. This has been requested by CFSAN and is also funded by CFSAN. As I mentioned, you have CBD and the primary metabolites are 7-hydroxy and 7-carboxy. The question is that this is -- if CBD or one of its metabolites, are they reproductive toxicants? So the way this is being addressed in vitro is to conduct in vitro incubations with testicular Leydig cells and Sertoli cells. This is being done.

Si and her collaborators will look at the morphology and the function of the Leydig cells and the Sertoli cells, and this is being done both in rodent cells and specifically mouse cells and in human cells. Where we stand on this project is, as I said, it's been funded. Si has started culturing mouse cells and both Leydig and Sertoli cells, and she's also trying to secure a supplier for human Leydig cells and Sertoli cells.

As I mentioned, there was concern about the fact that CBD is being added to cosmetics, and there's really no -- there's very little pharmacokinetic data to indicate what's happened. So in a study where Luisa Camacho will be

the PI, we're going to conduct a pharmacokinetic study where CBD is applied dermally to Sprague-Dawley rats. Then serial blood samples will be collected for a period of six days, and the way we'll make the measurements is by tandem mass spectrometry, HPLC tandem mass spectrometry, and we'll assess CBD, we'll assess 7-hydroxy-CBD, and we'll look at 7-carboxy-CBD.

Likewise, as I mentioned, there was concern when CDER reviewed the Epidiolex pharmacokinetic data, there was concern about the pharmacokinetic data. They are suggesting that additional data be obtained, and so in addition to the dermal study, we're also doing an oral study. Luisa Camacho again will be the PI on this. As indicated on the slide, this study was requested by CDER, and it's funded by sort of this FDA working group dealing with CBD.

Part of the impetus for this is -- and I believe Sherry Ferguson will talk about it tomorrow -- is there is a neurobiological/neurobehavioral study being conducted in the Division of Neurotoxicology, and yet there was not any pharmacokinetic -- say internal dosimetry measurements being made, and it was felt that this was necessary to be able to take the data that Sherry Ferguson is generating, you know, the neurotoxicological data or the behavioral data, and compare that to the pharmacokinetic data, because

then they could walk back and again look at the division with the Epidiolex.

The measurements will be made in exactly the same way. We'll do by tandem LC mass spectrometry.

The approach for this is we're going to -- there are going to be two types of exposures. There will be a gestational exposure where pregnant Sprague-Dawley rats will be dosed by gavage on gestational day 6 or from gestational day 6 through 17. Then there will also be a gestational -- there will be pregnant animals and then followed by treating the pups, and for this, the dams will be treated from gestational day 6 through delivery, and then starting on day 1, the pups will be dosed until either postnatal day 4 or postnatal day 21.

Again, blood samples will be collected. Tissues will be collected at terminal sacrifice, and the same CBD and its metabolites will be again assessed by LC tandem mass spectrometry.

Then the fourth study, again, Luisa is sort of a -- the study that Sherry Ferguson is conducting, there will be excess animals, and it was felt instead of just disposing of the animals they're not using, there could be some valuable measurements made on them, in particular with the aspect of looping back to the reproductive toxicology. So animals, excess animals from Sherry Ferguson's study,

will be used to assess whether or not there's any changes in the reproductive system on exposure to CBD.

The approach will be, again, this is taking animals from Sherry Ferguson's study, and then these are excess animals that she will not be using for particular measurements, and these, Luisa will use them. So animals will be dosed from gestation day 6 as well as postnatal day 21, and then they can be allowed to recover until postnatal day 180. At postnatal day 21 and 180, there will be full necropsies, and then the endpoints as you can see at the bottom of the slide will be a number of endpoints associated with this reproductive system, male reproductive system.

The third area I would like to talk about is COVID-19. This is obviously very new. It differs from the other areas, the tattoo area and the CBD area, in that these studies, the other studies were really requested by the product centers. These are studies that have, at least for the first two studies I'll talk about, were initiated by investigators at NCTR. So I don't need to talk about COVID-19 and what's happened for the first half of this year.

So really everyone's well aware there's no good treatment, at present time there's no vaccine unless you want to consider what the Russians are saying, and so we

thought in just thinking, investigators, as said in the beginning, in March, are there studies that we conduct that we could learn something about COVID-19, and we came up, investigators in the division came up with three projects. I would just briefly like to discuss each of these.

The first one is wastewater surveillance. This was mentioned briefly by Denise Hinton earlier this morning. The PI on this is Camila Silva. This is done in collaboration with -- Camila was trained in microbiology, and this is conducted in collaboration with investigators in the Division of Microbiology. More recently, she's developed -- these slides were put together about a month ago -- she has established collaboration with epidemiologists at the medical center in Little Rock.

As everyone's aware or should be aware, there is limited diagnostic capacity for COVID, for the coronavirus right now. Individuals do shed the virus in their feces. So a number of people are looking at this. This is not unique to Camila.

Then the other point is that wastewater surveillance is used for a lot of viruses listed at the bottom of the slide. So in early March or the middle of March, Camila said maybe we could look at the sewage treatment plants in Arkansas. So she contacted the sewage treatment authorities in Jefferson, in Pine Bluff, which is

where the first cases of COVID-19 were reported in Arkansas, and also the sewage treatment authorities in Little Rock, and has been able since the middle of April to obtain weekly samples and she's developed assays based upon on RT-PCR to assess the presence of the virus. I think the advantage of her approach over other approaches is she has really built in a lot of quality control, and we can't give you precise numbers at the moment, but it does appear that the levels at least these two areas have sort of maximized, the virus is maximized toward the end of July, which is sort of following the pattern that we're observing in Arkansas right now.

The second COVID-19 project I'd like to talk about is developing a flow cytometric approach to analyze COVID, SARS-CoV-2 antibodies in human plasma. The origin of this is that Jia-Long Fang, he's conducting a project with polyethylene glycols in 20, 40, and 60 kd, and he's developed a flow cytometric assay to look for antibodies to PEGs in human plasma. He's assayed about 200 samples now, and he can -- this assay allows him to quickly assess whether IgG, IgM, he's looked at it as a function of race, as a function of age, as a function of sex, and we thought that perhaps the same thing could be applied to looking for a coronavirus antibody in humans.

So the approach is that he is transfecting HEK 293 cells with fragments from either the spike protein or the nucleocapsid protein, and then he will then purify the proteins, couple it to latex beads, and then try to see whether or not antibodies recognize them, and then he'll develop a flow cytometric assay which hopefully he will be able to distinguish IgG, IgM and IgA, and then he has established collaborations again with investigators at the medical center in Little Rock to provide samples from patients who are positive for the virus.

Then the third project is the principal investigator is Annie Lumen. Annie Lumen is currently being funded by the Perinatal Health Center of Excellence to develop what she calls an agnostic, commercial-agnostic, PBPK modeling program. The idea behind her work is that when the reviewers at the Center for Drugs look at PBPK data using commercially available software, that's a black box and they don't know what's behind the code. So what she's doing with funding from the PHCE is to develop a program that the reviewers at the Center for Drugs will be able to understand what's behind the code. So she's currently funded, and now she wants to expand with additional funding to see whether or not this can be applied to agents that are being -- supposedly may be effective against the coronavirus.

So the approach is that they would -- by they, I mean Annie has two postdoctoral fellows who are working with them, and they have developed codes and they're gathering data from the literature, and then it will be applied to the drugs listed at the bottom of this slide.

This project is currently under review. The project with Jia-Long Fang is currently under review. If anybody is interested in looking at the protocol so they would understand what's going on and if they would have specific suggestions, we would greatly appreciate it, because it seems to me that this is the intent of having a review such as this. We have secured reviews or we are trying to secure reviews from the product centers on these particular projects, but if we had more input, I think it would just create better projects.

Lastly, this is my last slide, and there's a few what I will call challenges. The first one is what I've brought up before, and that, as Sherry Ferguson mentioned, it's tough hiring people at the moment because of the coronavirus. We have faced a very -- the last four years, we've faced a very serious situation that we are not allowed to have individuals in the centers who have not been in the United States for three years. I think this is quite detrimental to our scientific enterprise.

The same thing is -- we used to have a very active program where we would bring in visiting scientists for a period of maybe a couple weeks, a couple of months, and that's simply no longer possible, and I find it very disturbing, because I think science is multinational or international, and being restricted to being only able to use individuals in the United States is a terrible, terrible mistake. Not that you guys can do anything about it, but that's something I'd like to complain about.

Also, another serious situation we face is that we really are getting decreased funding from the National Toxicology Program. I think this is in large part because of a change in the leadership at the NTP. There has been a decreased emphasis on chronic bioassays, even though chronic bioassays are still used very heavily in the regulatory process by the FDA. We're trying to find alternate ways of funding projects that are being requested by the product centers, but it is a difficult time at the moment trying to figure out how we're going to do this.

And then a third thing, the CTP funds -- the Center for Tobacco Products -- funds the inhalation core here at NCTR. Up until about six months ago, the inhalation core was in a different division, and it was felt since they were doing research, that they should move to this division, and I welcome them. But every two weeks,

we meet with the CTP to discuss the projects, progress, and so forth, and it's been a learning curve for me to see how all this fits together to understand how the interaction with the CTP, and then it's also been difficult, because these are very labor-intensive studies to do these types of studies under the current situation where people really do need to stay apart for their own safety.

With that, I'll take any questions that you may have.

DR. ASCHNER: Thank you, Fred. I have a question, one of the COVID-19 projects. You mentioned the surveillance of sewer, I guess, or sewage, and I recall reading actually that the French have been doing so and in analyzing some samples, as early as September of 2019, they were able to find fragments of the virus in sewage in Paris. So I'm wondering if there's any similar programs that are ongoing in the United States and whether the plan -- obviously just doing it in Little Rock, it's going to be informative for Arkansas, and the city itself, but are there any plans to implement something like this?

DR. BELAND: Okay, the first point is Camila and I have talked about this, and it would be very nice if she could find sewage samples that, say, predated December, and she's looked around the country and has really not been able to find any. There is a national organization or co-

op, what you want to call it, where these exchange ideas as to how to go about it. But whereas when I talked about the study that Jia-Long is doing, he has blood samples that predate COVID. But as best we can tell, we have not been able to come up with any sewage samples that predate COVID.

DR. ASCHNER: Thank you. And you mentioned one of the challenges I guess that the division has, and NCTR as a whole, I mean, is it going to be possible for you to maintain the workforce that you have right now, or do you foresee that it's going to shrink over the next couple of years or three years, with all the funding issues that you're facing?

DR. BELAND: I don't think it is going to shrink. I think we're going to be creative. As far as finding postdoctoral fellows, Annie Lumen was able to recruit two postdoctoral fellows within the last year. Jeff Fisher and Darshan Mehta have been trying for a year to find a funded postdoctoral position and have not had any luck.

We're in discussions with the FDA chief scientist, is there a way that the FDA can step in, make up for the shortfall that we're having with the loss of the funding from the NTP, and the issue here is we lose our infrastructure; if we lose the ability, if we lose our pathology, if we lose animal care personnel, and then all of a sudden there's an issue facing the FDA. For instance,

we did the acrylamide study. That issue came up almost overnight, and we were able to put together chronic studies to deal with that. But if we lose all the infrastructure, it would be very difficult to put back together again.

DR. ASCHNER: Thank you. Are there any questions from the Scientific Advisory Board?

DR. COSENZA: This is Mary Ellen Cosenza. Great topics. I just have a question. You talked about the life stage of the pharmacodynamic PK modeling for the therapeutics for COVID-19. Have you looked at those models for the CBD work, since that seems to be potentially an issue as well?

DR. BELAND: The short answer is no, but I can have a discussion with Annie about that. We're always talking about what other drugs could we look at or what other exposures could we look at, and that certainly would be possible. But no, we have not discussed it.

DR. TROPSHA: This is Alex Tropsha. I also have a question about PBPK. I think it's potentially important studies, and kind of lacking in the scientific literature, because it looks like it's easier these days for people to get stuff into clinical trials than to study in depth. So I have noticed that you had on your list of drugs a few that have been discouraged. I wonder if you are trying to find explanations for low efficacy or low or unacceptable

low efficacy versus remdesivir that is still approved for emergency use, or what is the objective of looking at drugs that have been discouraged?

DR. BELAND: Okay, I assume you're talking about chloroquine and hydroxychloroquine. Okay, first of all, remember, this is a rapidly changing area, and these slides are a month old. I think -- and again, the proposal, Annie Lumen's proposal has not been funded. This is, when Bill Slikker mentioned this morning about a new funding cycle, this is for the new funding cycle. This proposal, this was a draft as of probably two months ago, and she's in the process of revising the proposal now. I think the submission date is at the beginning of September. But I agree with you, I don't know why; I see little point in working with chloroquine or hydroxychloroquine.

DR. TROPSHA: Well, except to understand the reasons for failures, which I think --

DR. BELAND: I'm not sure it was a failure. I'm not sure it was anything in the first place. The whole -- there was never any good data as best I could tell, that it was a political compound.

DR. STICE: Fred, great work, as usual. Just following up on the tattoo work and actually following up on the neurotox recommendations, where looking at multiple species, were there, depending on the results and what

results would trigger further studies, say, in a higher species such as a pig or nonhuman primate? What's your thoughts on that?

DR. BELAND: We do this, if we're talking about the tattoos, we work with -- Nakissa Sadrieh, the person we're working with, she's in the cosmetic division at CFSAN, she really would like minipigs. The trouble with minipigs is they are really not very small, and you're going to lose your Ns, the number of animals you could treat, really would be quite limited. So sort of the intent is we have convinced her that can we start with a mouse and then basically when we see that we can get with the mouse, then we could consider moving on to the minipig.

We have really never had any discussions about doing primates, and I guess really don't think that would be done at the present time. But the pig, we have discussed doing, and there have been studies here done in the past, done with tattooing and I think it was titanium dioxide, or it wasn't tattooing, it was dermal application of titanium dioxide done by Paul Howard a few years ago.

DR. STICE: I agree that the pig seems like a logical choice if the resources are available.

DR. BELAND: They are just -- well, they are not small. But we have talked about it. We want to start with

the mouse and then we'll see what we can get with the mouse and then we can go from there.

DR. STICE: The Yucatans are not small, but there are smaller ones. There's minipigs and then there are minipigs.

DR. BELAND: I'd like a micropig or a nanopig.

DR. STICE: Thank you.

DR. ASCHNER: Okay, let's go, thank you.

(Pause.)

DR. TONG: My name is Weida Tong. I'm a director of the Division of the Bioinformatics and Biostatistics. So our previous SAB meetings, actually it's about five months ago, I believe it's in March, so a lot of the information has already been exchanged by then. So what I'm going to do today slightly differently, and I'm still going to give a quick overview of who we are and what we do, and for the 2019, I'm going to highlight a few items which is not been covered in our previous meetings. However, I'm going to spend most of the time to talk about what are we up to nowadays in 2020 and where we go from here.

So the division has four branches. We have a bioinformatics branch and a biostatistics branch, scientific computing branch, and R2R branch, and the R2R stands for research to review and return, which by means is

to focus on in collaboration with other centers to support the regulatory applications. We have a little bit over 50 people, and their skill set is quite diverse, ranging from the conventional IT skill sets such as like a programming, software engineering, and database, all the way to the wet lab and experimentations. Forty percent of the division activities is in research, and 60 percent is in support.

Our mission is quite straightforward, and we wanted to make sure everything we do here has some relevance to the FDA regulatory process. So for that, we have a lot of the collaborative projects with other centers. So I'm going to mention some of them, as the presentation goes.

With respect to the research, we are continually working on the drug safety and toxicogenomics, endocrine disrupters, rare disease, and the drug repurposing, and the precision medicines. Now, the division, the primary focus on the bioinformatics and the biostatistics, and it is a difficult to take a footstep into our field without using some sort of the AI and the machine learning. So AI is sort of like bread and butter, and we use the techniques applied to the old research field and projects and we conduct it in our division.

We also have a specialized working group in our division called AIRForce, which stands for AI Research

Force. This group are involved in many different AI projects and more specifically they are working on the drug safety and applied AI for the food safety, in terms of the genomics, and we try to use the AI to identify the genomic biomarkers. Now a lot of the efforts, and we are working on, related to the FDA and documents. So I will mention some of these projects as my presentation goes.

For 2019, I'm going to talk about outreach activities and cross-center collaborations and in terms of the research accomplishments. As I said before, I need to point out a few things that I have not opportunity to talk about it in the last meetings. Also I'm going to provide some updates as well.

So for the outreach activities, I think there's one of the largest projects we are working on right now in this division is called SEQC2 Consortium and efforts. This project is close to completion now, and we have 18 manuscripts being generated and 10 of them already being submitted to the peer reviewed journals, and eight of them are going to be submitted by the end of this year. We also participated in European Long Range Initiatives to develop an omics data analysis framework for the regulatory applications, and we have a paper published in this area. Right now we are working with the OECD to incorporate this framework into the OECD, the guidelines.

We also participated in two challenges, and both are extremely interesting. One is organized by the FDA actually, is a part of the PrecisionFDA challenge, and this challenge is to address the issues of how to use the data analytical tools and the statistical methods to determine, to make a determination of the adverse events anomalies. So Dr. Leihong Wu from our division participated in this challenge, and he is one of the top performers for this challenge.

And we also participated another challenge which is organized by EPA, and this challenge is to evaluate the various machine learning and artificial intelligence methods to predict the androgen receptor binding activities. Dr. Huixiao Hong led the team from our division and participated in this efforts and his model is also on the top three performers.

Okay, so this is to summarize four collaborative projects with other centers. Of course we have more than these four, but those are the four projects we have been working on for a number of years now, and several of them already mentioned by the representative from other centers. On the left side, Dr. King mentioned about FDALabel as well as IND Smart Template. I'm just going to give a quick update for the FDALabel, and recently we conducted a survey

and we are very happy to find out that around 30 CDER reviewers to use the FDALabel and the databases.

For the Smart Template and right now we already have over 1,000 submissions being uploaded into the Smart Template. In average, we see around 40 submissions per month and the data being uploaded into our Smart Template. So one of the biggest milestone and we made in this year, and Dr. King has already mentioned, is in collaboration with OGD and Office of the Genetic Drugs, and both tools already now was developed to support the Office of New Drugs for their regulatory applications. So we are very happy to have this opportunity to work with the OGD to customize both tools in such a way that can be very useful for the OGD regulatory mission.

So this is the DASH project. DASH stands for Data Analysis Search Host. We have been working on this project for quite a long time now, and this project also close to completion, and we are in the process for production of this project. I just want to point out that we have over 800 applications have been captured in this database.

The last one I'd like to mention, because we are extremely excited about this project, not only this project is interesting, but also we have a strong support from the Office of Regulatory Affairs, on this project called ALIS,

which stands for Automated Laboratory Information System. The system is to manage laboratory samples and QC data, and the ORA has around 20 subject matter experts from the five ORA laboratories working with us, and right now we just completed the first model. It's a microbiology modules, which now is going to the production mode, and in 2020 we are working on the second module and of course the pesticide model.

With respect to research accomplishment in 2019, now I have been talking about the liver toxicity knowledgebase project for a number of years now. This is our longstanding project, and we are working on the liver toxicity and the knowledgebase for about 12 years in this division, and as a result, a lot of the scientists in our division get familiar with the issues related to the drug-induced liver injury, there are a number of satellite projects being generated and based on the knowledge we have from the liver toxicity knowledgebase, and one of the projects is funded by Office of Women's Health to develop a hepatotoxicity database for herbal and dietary supplements, and we just published two papers for this project.

Another project I think is also quite interesting is done by our statistician in our division in this division, they applied the Bayesian graphic models to integrate the high throughput in vitro assay data to

generate a comprehensive adverse outcome pathways to gain the underlying mechanisms of the drug induced liver injury. This paper was published in the Archives of Toxicology and received an award by the SOT Computational Toxicology Specialty Section in 2020.

So this is the two projects was mentioned by Dr. McMeekin from the ORA. This is a collaborative project with ORA using the AI, and one is a focus on species identification using image analysis with AI, and another one is applied machine learning and to assess the MS data set for the persistent organic pollutants. For each project, we generated two papers, and the two papers on this project and another two papers for the POPs, and we are just completed both projects this year.

All right, for 2020. The major efforts and we have made in 2020 is in the area of the artificial intelligence. Of course, and this is not only thing we do in this division, but clearly this is our major effort in these divisions. So here I am going to give you the three examples and one is related to COVID-19, another one is about toxicogenomics, and the last one, it's pretty interesting, it's about interpretability of the AI models.

Okay, so in April, we submitted a proposal to the FDA's Medical Countermeasure Initiatives about how we can use AI methodology in combination of the drug repositioning

principles to identify the market of the drugs that have a potential to treat COVID-19.

So this project was funded by the Medical Countermeasure Initiatives, and we have collaboratives from CDER and CDRH, as well as from the NCATS. So in the first part of this project is to construct the protein-protein interaction network for the humans, and from this network, we will be able to identify which particular proteins are regulated by the virus proteins and then which drugs will be able to inhibit on this protein in such way disrupt the regulations by the virus so that we would be able to identify the potential treatment.

There is a lot of discussions and progress has been made for this project, and we are already starting to prepare the first manuscript from the results and generate in the past few months.

Now this is the very interesting project, because we have been asked these questions for many years now. The question is can genomics data from the cancer cell lines predict the drug-induced liver injury? Now, cancer cell lines are really not much to do with the liver injury. Some of the cancer cell lines even come not even from the liver, not like the HepaRG, HepG2 in cell line, it is from the liver, for most of the cancer cell lines, not from the liver.

So from the biological point of view, this is a sort of out of the box, how that works, however there are several publications indicating that the cancer cell line contain the biology that are related to the drug safety. So we took the step on it, and basically and we took the genomics data for a large number of the drugs actually, and then we developed deep learning models to predict the drug-induced injury.

The results is extremely encouraging, and we already have the two papers, one already being accepted, another one just submitted, and see what the reviews going to say about these concepts.

So now I'm going to talk about AI interpretability, and this is the issue generates a lot of the discussions in the AI field, simply because the modeling organisms we use now tended to be much more complex and more like a black box. So the model might perform very well, but it's very difficult to interpret. Based on the common notion and the complex algorithm you use usually have good statistics but very poor explanations. But conversely, in some of the modeling techniques like logistic regression method or linear regression method, the model performance is not that well, however, and the model can be explained very well.

So now the question is what is the tradeoff? How much accuracy you are willing to pay for the explanation you can gain? Right? So we did quite extensive study using the several different endpoints, and such as the Tox21 bioassay predictions, using endpoints from the drug-induced liver injury, and the liver carcinogenicity as well. So what I'm going to do is the next slide, I'm only going to show the results from Tox21 bioassays.

So in this study, we have 68 endpoints. We applied seven different machine learning methods, and some methods, particularly these two methods, it's very simple methodology. It's a result easily to interpret, but other methodology is much more complex and difficult to interpret. We also used six different molecular representations, and again, some, it's easy to interpret, some easy to interpret and other it's a little bit difficult to interpret.

So without getting into any specific detail, you just simply look at this graph, you will see the results, it's a fairly comparable, and regardless how complex the modeling approach you use and how sophisticated the molecular representation method that we applied, the prediction accuracy, which shows on the y-axis, it's very much comparable. So we can draw the conclusion for the Tox21 bioassays, and actually we are more leaning towards

to choose the simple method and with the molecular representations that are easily to interpret.

But this is not always the case. Depends on what particular endpoints you are working with. So our really recommendation is if we are going to develop an AI model to predict a specific endpoint, a systematic evaluation, by including both simple and complex methodologies, it's really necessary to balance the statistical performance with the explainability.

Okay, I talk about the explainability. I also mentioned the term interpretability, and actually for many people, explainability and interpretability is used in an exchangeable way, but actually there is a difference in the AI field. Explainability, that means the model result you can explain in human terms. However, interpretability is trying to establish the causality, and which particular terms are driving the AI and the performance.

So a lot of the studies have been focused on the explainability and however we pick this up and on the causality assessment and using in the AI field, and more specifically and we developed an AI methodology called the DeepCausalAI, and actually when we developed this method, we are inspired by a challenged posted by FDA through the Fields(?) conference, and we are tasked to develop a method to address the opioid crisis. So the models we developed

called the DeepFAERS, and FAERS stands for FDA's Adverse Event Reporting System. Basically, we used the FAERS data by applying the AI and the deep learning methods to develop or to establish the complex relationship between the different -- apply the Bayesian approach to establish the causality. So right now, we are in the process of preparing the manuscript to summarize this research work.

So now I'm going to talk a little bit about the FDA documents and how we can use the artificial intelligence to address or to take advantage of the massive documents available in the FDA. So when I go out to give a presentation to people, always say the FDA has a lot of data, but my answer always, FDA do not have a lot of data. We have a lot of documents. Like FDA guidance documents is one type of document. The patient narratives is another kind of document. The drug labeling documents, which we developed FDALabel on it, and we have around 150,000 drug labeling documents, and each document have around 20 pages, 80 sections. It's a very large document.

If you are manually reading these documents, you are going to take a lot of the precious time to conduct the true review process. Of course, we also have a reading minutes, approve letters, so FDA has all kinds of different documents.

So now how the documents be used in FDA? This shows on the left side, and first, foremost, is to search the documents, and also, we wanted to extract the key data or information from the document, or we ask a specific question to the document, and try to get answers. So those tasks can be addressed using the natural language processing and we call it informational retrieval, named entity recognition, Q/A, and essentially, it's doing these sorts of works.

Another one is do the quality control. How are you going to remove the duplications and how to identify the outliers, we call it document comparison or anomaly detection in the natural language processing world. Also, writing a summary or understand the conclusion, based on the paragraph. So in the natural language processing, we call it automatic summarizations or sentiment analysis. So clearly, we use the modern AI methodologies, we will be able to address a lot of the reviewer task and the review in the FDA are facing.

So this is what exactly what we did, and the first product we are developing and it's not finished yet, and we are developing, it's a focus on document retrieval and topic segmentations, and the Q/A. The systems we call it called ASSIST, or Advanced Semantic Search and Indexing Systems for Text. So what we are trying to do is if you

have a pile of documents and we will be able to conduct a semantic search to pull out relevant documents, not just based on the keyword search. It's based on the semantic search.

Also we can use the semantic approach to group the document into the different categories so a reviewer will be able to quickly and drill down a subset of the document to review. So this can be applied in a variety of tasks, reviewer and for the regulatory applications, and one of the projects we are communicating with the CTP is called ASSIST4Tobacco, because we have a lot of the tobacco documents and normally take a long time for the reviewer to get into these tobacco documents to find the relevant document, and for review. So we are developing a project with the CTP right now.

If you are interested in for the drug-induced liver injury, this is what we are doing actually now. We can pull out all the literatures related to the drug-induced liver injury in the same locations. We can conduct the same and we can develop the same tools for the DILI in this search.

So you get the drill. If you have specific topics and you want to talk to us about how to do this, and I will be more than happy to discuss with you.

Now this slide summarizes the review process. When the sponsor sends a document to the FDA and usually these documents were stored in the electronic documentation rooms. So reviewer comes in and extracts this information from each document, put it into the template or spreadsheet, and then we bring them into the database for the future reference, and all for the future for supporting the future review process.

At this point, we have a number of projects and working with CDER mainly focused on the knowledge management side. Now we are starting to work on this side, and try to use the AI and to automatically extract the information, to fill out the template and the spreadsheet. We can use the text summarizations and other natural language processing methods.

So in the early days of the liver toxicity knowledgebase, when we worked on this project, the first task we did is read the drug labeling document, particularly reading the three sections called the boxed warning, warning and precautions, and adverse reactions. Based on these informations, we will be able to make a determination which drugs are of most concern in terms of the liver injury, which one is less concern, which one has no concern.

Now we not only just did it once. Actually we have two scientists did it independently, and we combined this information and make this call. So right now, we are developed the AI, and we ask the question whether AI will be able to recapitulate the expert's knowledge, based on these three sections to recreate DILI classification which was done by the experts.

Okay, so just outline two particular projects, and we are of interest to work on, and one is about systems toxicology. Now, we have been working on the TG-GATEs project for a long time. The TG-GATEs dataset, it's very large and it's generated by a Japanese consortium. They took 10 years of $50 million to generate this data. So without getting into any specific detail, I just want to point out two datasets and those are the in vivo assays and they have the histopathology data, was generated actually, image was generated for the liver, and the total they have 12,000 images. So we showed these images to the pathologists and asked them whether the annotations made by Japanese group is accurate or not, and you may guess it, and they come back very, very different opinion.

So what are we doing right now, we are going to use the AI to systematically read all these 12,000 images, but most importantly, for behind every image, there is gene expression data that's available, and better than that, we

also generate the microRNA data. So what we really wanted to do is to conduct the integrated analysis of the imaging and the image data, with the gene expression data from the microarray, and the microRNA data from the next generation sequencing.

So this is my last slides, and just talk a little bit about some of work we have and where we are heading for the AI. In my mind, AI, if we are going to apply the AI in the regulatory science, we need to have a rigorous evaluation of the AI. Just like the drug, and go to the human use, we need to conduct the clinical trials.

So we developed so-called AI TRIAL principles, and the key, transparency -- which stands for transparency, reproducibility, interpretability, applicability, liability. Now in terms of the transparency, in most of the journal right now it's required you deposit the data and even code into the public repository. So the transparency actually is not an issue. Once you have the code, you have the data, you can repeat it. But for the AI, it's much, much more than that in terms of the transparency, particularly when we apply AI in the regulatory setting. We need to understand why an AI model behave in a certain way.

Now in terms of the reproducibility, again, if we replicate a study, it's very straightforward and easy.

However, some AI model, you cannot even replicate it. For example, like a BERT model developed by Google. They take years, humongous, and computational resource to generate these models. I think only Google can reproduce their models. Okay?

So replicability and repeatability is important, but sometimes it's not feasible in the field of AI. So what we really needed to address of the reproducibility is how can AI findings be trusted, and we need to address these particular issues to make sure the AI models are reproducible.

Now I already talk about interpretability, and we certainly need to look beyond explainability, and we need to look at causality, and applicability for any people's working in the field of AI, and they know if we develop a model, we really need to have a good sense of the context of the use of these models. It's not for every AI model will be applicable in every situation. It's more important than the statistical performance. It's how we can define the fit for purpose of the model applications.

So lastly is the liability. Now this is not my field, and this is really getting to the ethics and other policymaking. Fortunately, in FDA, we do have people working in this area, and we are going to work with them to address the liability issues.

So those are the principles then to guide us in the future development of the AI in my division. I'm going to stop here and if you have any questions.

DR. ASCHNER: Thank you, Dr. Tong, for a very interesting presentation.

DR. TROPSHA: Weida, thank you for a great presentation, really very comprehensive collection of thoughts and projects. I have one kind of more organizational and a scientific question. Organizationally, can you comment on the dynamics of your interaction with other centers? I see that you develop a lot of tools and databases. Was not clear whether this is initiated by you and then kind of shopped to users, or whether users from other centers come to you. Can you clarify that for us?

DR. TONG: Alex, great questions. Yes, and for example, we developed ASSIST for tobacco, this is -- we are working with CTP right now, and I'm not going to say we are already starting to work with them, we are communicating with them, and we jointly developed protocols, and now the protocols is in upper management for approval. If everything goes well, we will start to implement the ASSIST4Tobacco, that particular tools for the CTP use. Some other AI methods we developed here, for example, that we called the DeepReviewer. This tool is already being

applied to the data, and stored in the DASH as well as for the IND Smart Template.

So yes, it's not all of the tools have been used by the reviewers, but some are already being used by reviewers.

DR. TROPSHA: I love the abbreviation of the screen, and all the work you are doing with AI, but I think even your studies show, and the emerging papers, showing that in many cases, the AI methods don't provide any advantage, statistically speaking, over much simpler methods. So what's the justification of pushing in this direction?

DR. TONG: Very good questions. First of all, I am a really big fan to conduct a systematic analysis, not just pick one method, for the strategy. If you have an endpoint and definitely, I will suggest a study with the simple method, and normally the simple method provide a much better explainability. Now, with that said, and the deep learning does show several advantages, particularly when the data is diverse and from the different -- we normally call it from the heterogeneous fields. Let's just say you have genomics data, you have a proteomics data, you have a metabolomics data, and it is a really difficult to integrate all of these informations into the single platforms to develop models, predictive models.

So the simple method might not be able to handle this type of tasks. So the AI can play a major role in the systems toxicology and the systems biology, whatever you call it. However, when we develop these kind of tools and we really need to keep in mind, if these tools or models are going to be used in the regulatory applications, we need to follow into these rules on the screen and we need to make sure the transparency, reproducibility, interpretability, applicability, and of course the liability as well.

DR. RAMOS: Weida, I was commenting on the fact that you always manage to capture my imagination and to make me think far beyond what you're thinking. So I enjoy your presentation very much. I have a couple of questions, and then a request.

My first question relates to your commentary about the toxicogenomics DILI project, in which you took cancer cell lines and then try to predict liver toxicity. Can you provide some more granularity to what you actually did in that particular study, and let me tell you what I'm thinking of exactly. A comment that was made before, which I think is very important, is the fact that oftentimes cancer cell lines lose metabolic competencies, and in the case of liver, oftentimes liver injury is connected to metabolic activation and detoxification pathways. So I'm

curious to see the extent to which your model was biased by the selection of chemicals that you actually chose for testing that model.

Go ahead and get that one, and then I'll ask you the second one.

DR. TONG: Great questions. What we see, actually we use the deep learning method and we try to establish the causality, so what we found and some of the key pathways associated with the drug induced liver injury, that's captured in the cancer cell lines. Not all of them, particularly metabolic competency, right? So that means the models we develop not be able to predict everything 100 percent correctly, but so does the animal model. So our performance is much better than animal models.

And then I would like to take a step back, and when we talk about the predictive models, it's not about capturing every moving part with the biological significance. It's sometimes you only need to capture several important biology will be able to provide the good predictions. So this is what we observed from the model, and we developed, using the deep learning method.

DR. RAMOS: I hear you. Obviously we'd have to chat offline to go into the nitty-gritty details, but I would like to actually get a copy of the paper and read it,

because I think I'm curious to see how chemical selection could have biased the results that you actually generated.

Then my second question relates to the AI causality model that you talked about. Very intriguing concept. You know, especially in the light of when you think of AI, you normally have a bias towards predictive analysis, and in the case of -- when you think of causation, you're really reverse engineering a problem. So it's an interesting sort of flip to the idea.

My question related to that particular model is how did you validate that model? Did you use the opioid case to validate, or did you actually take a well-known relationship, if you would, of causation to actually run it through the model and then see how it functioned?

DR. TONG: Great question. I said it's the opioid crisis, and actually at the end of the day, we did the liver injury. So the causality model is, because we know drug-induced liver injury and we know what's really going on or what kind of drugs are going to cause liver injury. So what we did is we say, okay, we observed the hepatic failure and which drugs cause hepatic failure, and using our causality, AI models, and what we found is acetaminophen. And then the next question we asked, well, which age group of the patients take acetaminophen and have

hepatic failure, and again, we found that the specific age group.

And then we asked the question, what particular dose of the acetaminophen these patients would take in the cause of hepatic failure, and we found it's over 100 milligrams dose is exactly consistent what we understand about the drug-induced liver injury. So we almost finished that draft manuscript, and I would be very much happy to share the manuscript with you.

DR. RAMOS: I will look forward to reading it. I'll stop there and catch up with you at some other time.

DR. ASCHNER: Thank you, Ken, and thank you, Dr. Tong. Thank you very much for again very interesting presentation.

We are moving on to our last presentation. It's from the Division of Genetic and Molecular Toxicology, and it's going to be presented by Bob Heflich. Bob, please go ahead.

DR. HEFLICH: Hello, everyone.

My job for the next 30 minutes is to fill you in on the Division of Genetic and Molecular Toxicology. I serve as the DGMT division director and Mugimane Manjanatha is our deputy director. I can see there are people who are still on the line here, and we seem to be getting a higher percentage of genetic toxicologists. So I think that's a

good sign people are actually interested in what we're doing.

Here is the division. We have 34 staff members at present. Among the government positions, we have 15 PIs, research scientists and visiting scientists, and 10 support scientists, plus our ORISE postdocs, give us a total of 32 scientists, with two administrative support personnel.

Our mission is on this slide. Our mission is to improve public health by providing FDA with the expertise, tools, and approaches necessary for the comprehensive assessment of genetic risk. Our research goals have been to respond to agency needs for chemical-specific data. Recent examples are tobacco products, nanomaterials, and drug impurities. Also, to maintain leadership in regulatory assay development. Some that we have worked on over the years are listed in the slide here. Lastly, to establish new paradigms for regulatory decision-making. In other words, advancing regulatory science as it relates to genetic toxicology.

I've attempted to divide up our research strategies as follows: first, to engage FDA and other scientists and stakeholders to set our research priorities. Secondly, I've sort of made two research themes out of our laboratory research, first of all, to develop better

biological models, and we have recently put a lot of effort into in vitro tissue models, and secondly, to develop more comprehensive ways of monitoring genetic risk, especially utilizing error-corrected next-generation sequencing approaches.

I'm going to concentrate on the second theme and the last bullet here. Tune in next year to hear about what we're doing with in vitro tissue models. Actually it may have been a good choice, since Dr. Slikker and Dr. Braunstein, Dr. Tan and Dr. van Bemmel, have already mentioned this work. But you need to hear more about the genetic analysis stuff. No one has mentioned that yet.

This next slide summarizes some of our outreach activities. The top lists a number of examples of collaborative partnerships with other NCTR, FDA product centers; I left off CVM here. This is an old version of the slides. Other government agencies in the United States and abroad, and academic institutions and other organizations. Below lists some instances where we have taken the lead in various outside activities, including committee leads and various international organizations like HESI, Health and Environmental Sciences Institute, IWGT, which is the International Workgroup Genotoxicity Technology Methods, and the OECD. We have had a president and two secretaries of EMGS in recent years.

Not listed on this slide, we also count among us the current editor of Mutation Research, which is a major journal, specialty journal, in the field of genetic toxicology. I'm a past editor-in-chief of Environmental and Molecular Mutagenesis.

So as I said, for the bulk of my talk, I would like to tell you about some of our ongoing projects that have made progress recently by employing error-corrected next-generation sequencing methods, techniques that we have invested heavily in over the last few years, and which we feel may be transformative technology for the field of genetic toxicology. So I've listed four projects on this slide, and I'm going to tell you a little bit, tell you more about the last three that are in bold.

The first bullet refers to a method that I will not have time to go over in detail, but Dr. Braunstein mentioned it in her presentation, which is comparison of genomes of single-cell clones generated from mutagenized and non-mutagenized animals, mammalian cells, or bacteria, depending on what you're looking at. This approach has been used by Javier Revollo, Tao Chen, and their colleagues to evaluate the mutagenicity of nanomaterials in mammalian cells, germ cell mutations in C. elegans, and the off-target effects of gene editing. The plan is to use this

approach as part of our Pig-a studies in the upcoming year. I'll mention something about that later.

I can tell you something about how this works, this clonal comparison method works, if you want in the Q&A session.

I am going to spend some time describing recent results with the next three EC-NGS methods listed in bold. CarcSeq, which is a method for analyzing driver mutations. This is work conducted by Barbara Parsons, Meagan Myers, Kelly Harris, and Karen McKim. Duplex sequencing for measuring mutations in highly differentiated in vitro tissue models. This is work being conducted by Yiying Wang, Xuefei Cao, Bo Mittlestaedt, and Rebecca Wynne.

And MAML, which is an acronym that stands for mutational analysis by multiplex libraries. This is an approach developed by Javier Revollo again, Vasily Dobrovolsky, and Azra Dad. This has overcome some knotty problems with getting regulatory acceptance of the Pig-a assay, which I'll mention later.

So probably some of you are not familiar with the problems with detecting rare events with next-generation sequencing. I put in this slide here to explain what this is all about. I guess everyone is aware of the power of NGS to generate lots of DNA sequence information. The approach involves sequencing a target over and over again

for hundreds and thousands of times. So you can imagine that if you have a rare variant sequence, like a somatic cell mutation and a frequency of, let's say, one in a million wildtype sequences, and you sequence that part of the genome where the variant is located enough times, some of the output data will be the sequence of the variant along with a lot of wildtype sequence. The mutant frequency can be calculated. Now you can see the problem with doing this is that the standard NGS techniques used for sequencing genomes have an error rate of about 1 percent, meaning that for every 100 patients there is a sequence, NGS will make a mistake.

If you look at this upper graph here, this illustrates this; if you look at this output from the stretch of DNA, the orange -- I have stolen these figures from Jesse Salk and Kennedy from a paper they published -- you can see that this error rate sort of is a tremendous background obscuring any rare event.

Now, if you want to find a consensus sequence, knowing that you only want a predominant genome, you can ignore all this noise. However, how do you find a rare genomic variant in this sea of errors? What you need is error-corrected NGS to locate the real variants, and that's shown in the bottom of the graph. Clever people, including some of the DGMT, have devised various ways of doing this.

I will call these methods error-corrected, because that is what everyone else calls them. But actually they seem to me to be more correctly involved error avoidance. I hope I can explain some of this to you in the next couple of slides.

Let's start with CarcSeq here. Barbara Parsons and her crew are currently developing panels of cancer driver mutation targets to serve as biomarkers of effect for cancer. The goal here is to incorporate this analysis into a repeat dose rodent study, maybe a 28-day or 6-month repeat dose study, to be able to identify rodent and hopefully human carcinogens, using expansions of cancer driver mutations, all without counting tumors.

You're probably aware that CDMs by definition have a growth promoting phenotype, and as outlined in this diagram, where they show up as biomarkers of effect, cause cooperating clones and cells to expand into what eventually forms a tumor. So one key piece of information is that CDMs by their nature amplify the mutation with time, helping to overcome the NGS error rate. You can see that applying some additional methods and rules, and Barbara developed an EC-NGS method to efficiently synch with dozens of cancer driver mutations, all at once. One interesting aspect of this is that the CDM analysis may be able to distinguish between carcinogens with a genotoxic mode of

action; that is, induce cancer driver mutations de novo in carcinogens with a nongenotoxic mode of action. That is, that cause expansions of preexisting cancer driver mutations. At least, we hope that will be the case.

So on the left is a panel they are testing out for lung and breast tumors that contains about 30 mutation hotspots, evaluated about half that many amplicons. The idea is to use CDM specific to the tumor type. These hotspots are identified from cancer databases, and especially those that are conserved in human and rodent tumors. This is the rationale for hoping that the rodents can be used for better prediction of human responses.

If you notice, some of these hotspots are much more prevalent in one tissue than they are the other. So the expectation is that CDM specs(?) will be tissue specific. The text on the right outlines two studies that have been conducted using this panel. In one study they evaluated normal and tumor tissue samples from human lung and breast, and another they analyzed tissue from three rat species with different spontaneous incidences of mammary neoplasms.

This slide sort of summarizes the findings. By itself, CarcSeq has a sensitivity of about 1 in 10,000. Not shown here, the tumor CDM frequencies it produces correlate very well to frequencies in much more laborious

methods that this group has used in the past. In the human tissue study, they found that expected spectral analyzing nine normal using breast samples and nine normal lung samples in comparison with nine to ten human breast and lung tumor samples. An interesting tidbit is that the CDM frequencies increase with age, and that's shown over here, as you might as you expect, the sum of the lung specific CDM frequencies in human breast tissue as a function of age is shown here.

In the rat mammary CDM study, they found a correlation between baseline CDM frequencies and tumor susceptibility. So these data seem very encouraging, and I will say something about future plans in just a bit.

So I will say something about our in vitro models. We have done a lot of work starting about 10 years ago with adapting in vitro and organotypic tissue models and making toxicological evaluations. This slide shows what one of our best developed models looks like in cross-section. This is the air-liquid interface, human airway model.

If you look on the left, the model has an air interface and I'll try to -- this side here. On the apical side, you can see where the cilia are on the apical side, and they sit -- the model sits in media on the basal side, and I'll show that on the next slide. It might be a little

clearer what these things look like. We have a lot of equipment, very sophisticated equipment and methods to expose these cultures to vapor aerosols and cigarette smoke, and there are a lot of disease-relevant physiological endpoints that can be monitored with these cultures.

But one thing that's eluded us, however, is how to measure genetox endpoints in the cultures. You can see in the panel on the right that the cultures contain precursor or basal cells and that's the dark stain p63 positive cells, and dividing cells, the Ki-67 positive cells that maintain the cultures as a steady state. Once they differentiate into this structure, they essentially stay steady over a period of months, three months to six months, which you can do experiments with. So there are some dividing cells in these cultures, but they are very few of these progenitor cells, making the fixation and expression of genetic damage hard to detect.

So what we have done is conducted a little experiment recently and we treated these in vitro cultures as if they are in vivo tissue and performed a 28-day subacute study like you would if you were performing a gene mutation assay in a transgenic rodent like Big Blue, slide shows the treatment and sampling schedule we employed for a proof of principle study conducted with ethyl

methanesulfonate, which is a direct acting ethylating agent. We also did Comet analysis and toxicity assays, and attempting to do micronucleus assays with these cultures.

So you can see on the bottom here what the cultures look like when they're actually being cultured in a dish. The tissue sits in this little insert, which has a membrane that allows feeding from the bottom, with media, so the air interface remains open to the atmosphere.

So these cells from these highly differentiated cultures wouldn't clone very well. So we can't select for HPRT or TK mutants, as is done in conventional mammalian cell gene mutation assays. To make single cells will mess up their cell surface. So analyzing for Pig-a mutations is problematic, although perhaps not impossible.

But in order to evaluate gene mutations in these cultures, we use what is called duplex sequencing, and that's shown in this slide, which labels the duplex strands of DNA differently with linkers. Over here, you can see the two strands plus and minus strands, call them, one with these two linkers, one a purple one and one a green one, to differentiate them.

This is another figure I borrowed from Jesse Salk, who apparently isn't on the line, so I don't have to worry about it. But anyway, so after you sequence the individual strands, you can identify whether or not the

data are derived from the plus or the minus strands. As you can see in the middle here, real mutations will be in both strands as the consensus, whereas the red bases indicate where errors were induced randomly in one strand or the other. This conceptually simple trick for identifying real sequence from random changes is able to pick out one mutation about 10^8 bases at close to 100 percent accuracy. Not easy to do, but tremendously powerful technique.

And it worked for us with our tissue model. Here is a proof of principle study showing a dose response increase in gene mutation after 28 days of treatment with nontoxic doses of EMS, two samples each shown here from the control, middle dose, and a high dose, plus this is a control sequence that's added for the assay. We have more data since this slide was finalized a month ago showing two additional lower(?)responses, producing a nice dose response.

The bottom line is that these techniques, these error-corrected NGS techniques, like CarcSeq and duplex sequencing, open up incredible possibilities for doing genetic analysis. The risk of mutational damage can now be evaluated in any tissue in any sequence, including working at the whole genome to get a total genomic load, but of

course those kinds of experiments are tremendously expensive.

Okay, for our third accomplishment, I would like to say something about the Pig-a assay, which also has an EC-NGS angle. As you are probably aware, the Pig-a assay measures the presence or absence of the cell surface anchor showed here, highlighted in blue and yellow, by analyzing microliters of peripheral blood using high throughput flow cytometry. The regulatory assay is an in vivo assay for gene mutation that can be conducted in mice and rats. Loss of the anchor means that the cell has a Pig-a mutation, and the regulatory assay, Pig-a mutation is measured using peripheral blood erythrocytes, and as you probably know, there's millions of peripheral red blood cells, peripheral blood erythrocytes, in drops of blood. So it requires very little sample to do the assay.

So we've been working on getting regulatory acceptance of this assay since the mid-2000s. Some of the steps along the way are listed in the slide. About 2010, we decided that the best route was by developing an OECD test guideline for the assay, which you're probably aware is the gold standard for international regulatory acceptance of genetox assays. The assay is recommended, even though it doesn't have an OECD test guideline; it's

recommended for use in several guidances, including for drug impurities. So it has relevance to FDA testing needs.

In April, we achieved a major milestone in this long road, the milestone is shown in bold here, when OECD accepted the large amount of documentation we had developed on the assay and its validation and gave us the go-ahead to prepare a test guideline.

Although we'd like to, we can't take full credit for getting where we are on this. This has been a result of a major collaborative effort from people from around the world, scientists from North America, across multiple countries in the EU, Japan and Korea, and China, and we're currently working on developing with them a consensus draft test guideline working through HESI, and next steps will work with an OECD expert working group that's been formed for this. Hopefully, the test guideline will be ready for review by the WNT at the next meeting in April of 2021.

So now back to error-corrected NGS. One of the challenges in validating the assay was in determining that the assay actually measures mutations in the Pig-a gene, which is no mean feat when the cell is used for the assay. Red blood cells don't have DNA or RNA. So Javier and his team went to the cells where the Pig-a mutations likely occur, bone marrow erythroid precursor cells, and sorted out the potential mutants and sequenced them using a method

they called MAML. If you ask me, in the question and answer period, I'll tell you exactly how it works. It's very clever.

Using MAML, we developed Pig-a mutational spectra for rat erythroid cells using four mutagens. This work was all supported by CDER over the past three years.

So to take the big picture of the sequencing work, this slide is from the OECD submission that shows that this revision of mutations that we found in the Pig-a gene indicating that the assay has a reasonably large target for mutations. Note that the mutations were found from the beginning to the end of the coding sequences, and very few in noncoding sequence, and those that were generally were in consensus splice sites. So the link for finding all this in the rest of the documentation is up here and is three files totaling about 400 pages.

Different compounds also produce unique mutational spectra consistent with the types of DNA damage that they produce. This figure compares mutations induced by procarbazine, which generally makes adducts with dAs, with those induced N-propyl-N-nitrosourea, which generally produces adducts with dTs.

The data is the mutations are expressed as the sequence change on the non-transcribed DNA strand, while the two compounds, both make adducts at AT base pairs,

strand-specific DNA repair causes the damage on the transcribed strand to be repaired, leaving the damage on the non-transcribed strand. This is all very pleasing to me, because it shows the intervention of biology and chemistry of the adducts in resulting mutations. So very nice data.

Okay, now let's say a little more about future plans. I don't think we will change the major direction of DGMT very much. I think we have developed niches for ourselves in which we're doing work that has impact not only for FDA but for the regulatory science in general. So we're continuing doing what FDA needs us to do regarding standard genetox methods, and I haven't said anything about that, but we do support the genetic FDA product centers in doing a lot of the standard assays when they ask us to.

Last, emphasizing the development of in vitro models as well as developing error-corrected NGS methods and applications. Some of the areas that we have just started or plan to start with regard to in vitro models are listed on this slide. Note that because I have listed them here doesn't guarantee they will be in the accomplishment pile next year. Some of these projects will likely outlive me. What we are doing here is seeking your counsel on them before we get in too deeply.

First, we will continue adapting genetic toxicology to our in vitro tissue models. I've listed some possibilities here, but most especially developing a micronucleus assay for the airway model to go with the gene mutation endpoint. This will address a proposal for one of the product centers that was made recently that it would be nice to be able to address in a positive manner.

Secondly, I'd like to develop -- we would like to develop complementary rodent and human models. Often the reference data for validating the performance of in vitro assays are in rodents. But in the airway model I showed you before, it's a human model, and also what we want to ultimately do is to evaluate risk to humans. So ideally, we need models from humans and lab animals, both to validate the assays and determine what the models can tell us about human risk, and that's doable with many of these models.

Thirdly, and this was mentioned by the previous speakers, we have done a bit of work with in vitro mammalian cell infection models, specifically a Bordetella pertussis infection model in airway cells, in Zika virus infection model in testicular organoids, and now work is poised for coronavirus studies in the near future. I think Dr. Slikker presented a proposal for a coronavirus study

using human airway ALI model. We intend continuing this work on these infection models.

Next, one thing that has to be done is to standardize the in vitro models and endpoint evaluations that are going to be used for making regulatory assessments. I realize this is a general problem or general issue with using any sort of new method for regulatory assessments, and Suzy Fitzpatrick mentioned that in relation to her work.

We are moving in this direction with support from the NTP. They are interested in the ALI airway model, and with the collaboration with IIVS, that's the Institute for In Vitro Sciences, to develop standards for this model. There's also a collaborator that we have CDRH that's interested in developing the model as a biomarker for chemical sterilant evaluation, inhalation of chemical sterilants, and he's thinking of using the CDRH biomarker qualification program to do this.

Finally, we recognize the natural progression of work with these in vitro tissue models is to reduce some of the reliance placed on in vivo work for making regulatory safety assessments, and again, Dr. Slikker mentioned this in his overview of NCTR. A division member is beginning to get some training in computational modeling, and in vitro to in vivo extrapolation, but we are just starting the

effort and hopefully a lot more resources can be brought to bear for making progress in this area.

As far as the genetic analysis is concerned, we're continuing work on the Pig-a assay this coming year to show that the mutations detected in the Pig-a gene are representative of mutational load in entire genome, and this work has been supported by CDER. That's directly testing how well Pig-a works as a reporter of in vivo mutation. This will be done using the clonal comparison EC-NGS method that I mentioned previously.

Secondly, most of our current EC-NGS studies rely on Illumina sequencing, generate sequences of only a couple hundred bases, meaning that large genetic changes may be hard to detect. We would like to explore the potential benefits of a new instrument that we have recently acquired, the PacBio Sequel II, which I may have misspelled here. Yes, it's an E, not an I. Which should increase sequence rates by a factor of 10.

Besides developing CDM panels, the third point, besides developing CDM panels for common tumors in humans and rodents, it's time to do some test cases. In other words, some proof of principle studies using treatment of rodents with genotoxic and suspected nongenotoxic carcinogens to test how well the paradigms for detecting these two types of carcinogens work, a collaborative

project is being put together with the CDER scientists, and Barbara Parsons is in charge of that.

Also, we would like to adapt the CDM analysis to some of our organotypic models to perhaps get more relevant biomarker of cancer using a more relevant human cell system. So we have, for instance, we have that airway system wouldn't be interesting not just measure reporter mutations but to look for expansions in cancer driver mutations in this model, if indeed our interest is in lung cancer, for instance.

So asking for a little feedback, two general points of emphasis are repeated in this first bullet. Next year, I'll say a little more about in vitro tissue model accomplishments, and to add a little more fuel to the fire, Donna asked that we present our futuristic plans, and I interpreted this kind of broadly. This is kind of a back to the future I guess in my way of thinking.

It's clear that these kind of goals will take a lot of effort to make an impact on the world of regulatory science, but my future goal is to develop mutation as a true toxicological endpoint for doing quantitative risk assessment. In other words, using mutation per se as a risk factor rather than, for instance, as a biomarker for agents as carcinogens and noncarcinogens, as we do currently. This is a back to the future thing, although

it's has been around for about 100 years, starting with the ideas of Hermann Muller, who later won the Nobel Prize for his work with x-rays in the 1940s, I believe, with fruit flies.

But we sort of got off track building this when the Ames test was developed, and hazard IDs that followed were so incredibly important to regulatory science. But I think with the tools that are being developed today that we can comprehensively identify alterations to the genome at very low frequencies, that goal is becoming ever more achievable.

So in this last slide, so for those of you who are interested in this idea, I would recommend taking a look at this commentary that we put together to honor the 50th anniversary of the Environmental Mutagen Society, and lest you think that this is a fringe idea, the authors of this paper include the current and preceding three editors in chief of the Society's journal, Environmental and Molecular Mutagenesis. Also included are senior genetic toxicologists working with the NTP and Tox21 programs, and two Europeans, one of whom works for Roche whose experience with a drug contaminant about 15 years ago spurred rebirth of this idea.

I told my coauthors that I would be giving them a little advertisement during this talk. So I hope some of them tuned in. I think they have, actually.

This is where I stop and leave it there and entertain any questions you may have. Thank you.

DR. ASCHNER: Thank you, Bob. Thank you for a very nice presentation. Okay, are there any questions? I know it's late in the day. We've been on the phone 9 hours and 17 minutes actually, to be exact. Does anybody have any questions?

What do you see as the challenges for the division in the next year or couple years?

DR. HEFLICH: Well, I personally think that of the 40 years I've spent at NCTR, some of the most exciting things are happening right now. I admit to being biased, and these are the sort of things that I've thought about for a long time, but I think this movement into using mutation in a more sophisticated manner and these tools that we have now to analyze mutations and things other than cell lines and reporter genes are just so powerful and open up so many new possibilities that the challenge is convincing other people that maybe doing Ames test and the mouse lymphoma assay may not be the end of the story for genetic toxicology, that there's a whole new world out

there that they should be thinking about and seeing the potential advances that could occur in this field.

So as I say, I'm not sure -- regulatory agencies are by their very nature very conservative sort of organizations, and our tools that we've developed over the years are very useful to them. So these sort of new ideas are something that will take a while to take root and how to do that in sort of a constructive manner and incorporate this into regulatory science will be a real challenge in the future, and that's something that I like to think about and I like to have -- I like for our division to have a part in it.

MICHAELKAWCZYNSKI: Ken, you might have to hit #5. Sherry is typing a question.

There we go, Ken. All right, Ken, take it away.

DR. RAMOS: Bob, I enjoyed your talk this afternoon. So I have a couple of questions. For the CarcSeq methodology that you introduced us to, I think I read on the slide that there's a bias towards coding sequence in the methodology, but then later on you made reference that you could actually apply this technology and this approach to whole genome sequencing. So I'm curious to see which of the two is it, and then depending on what you answer, I'll ask another question.

DR. HEFLICH: I didn't mean to give that impression. The four things I mentioned as sort of applications of so-called error-corrected NGS are sort of fit for purpose approaches, and they're different. The CarcSeq actually targets those 30 -- at least for in lung tumors -- those 30 hotspot cancer driver mutations. So it's very specific, and I think the number of bases that are actually interrogated is a little over 1,000. So it's not a very big target.

DR. RAMOS: I see. Then, so you didn't mean to make reference then to whole genome sequencing.

DR. HEFLICH: Oh, not in that context, no. Actually, we have tried some of these other methods to do hotspot cancer driver mutations analysis, and because they look at more sequence, they're quite a bit less efficient at doing that. So you get a lot of other things, along with your driver mutations, that you don't really want to look at. So the CarcSeq has an advantage in sort of targeting these particular mutations that are relevant to cancer.

DR. RAMOS: But to your point about the utility of the assay of a mutational-based assay, you know, for broad screening of chemicals, then you run into the challenge that you are neglecting a sea of other genomic changes that are equally important in cancer initiation and progression

that would not be covered by a targeted approach. I'm thinking particularly for noncoding sequence and copy number variants. So what are your thoughts on that?

DR. HEFLICH: I think you are totally correct. We're looking at the low-hanging fruit here by looking at target mutations, and there's a good -- there's a lot of information that would support that that would be the place you would start, but if you're -- especially if you're interested in something other than cancer, you certainly wouldn't use a method like this. You'd use a more generalized method, and even the way you -- the kinds of tissues you looked at and the treatment regimens that you used could be completely different.

FDA tends to use genetic toxicology data to regulate on cancer. So that's one of the places we start, and in fact, doing a cancer bioassay that actually go to tumors is a tremendously expensive proposition and something that's less and less done these days. So we thought this might be a way of sort of providing something that would be immediately or in the short term relatively useful for FDA in making their regulatory decisions. So that was the idea there.

But there are a lot of other kinds of genetic toxicology or mutational events that are important to human health, and I guess I'm preaching to the choir here, but I

mean, if germ cell mutation is somewhat given a short shrift in the FDA, and that could be something that we could look at using these models. In fact, one of our investigators has a nice C. elegans model where we can look at germ cell mutation using whole genome sequencing of the worm offspring. So sort of a clonal expansion of a worm in other words to look for germ cell mutations. So that's sort of a neat application, and of course with C. elegans, you have both male and female germ cells you can hit, depending on your timing of your treatment.

DR. ASCHNER: Okay, thank you. I don't see any more hands up. So we're running a few minutes late as well. I think I want to thank Michael for running this very smoothly. We'll forgive you for your batteries running out. It was wonderful. It was wonderful. I think we can talk about it tomorrow, about the format and so forth, but I think it went as well as we could have expected; other than the interference in the morning, everything went very smoothly. So thank you, Michael.

I want to thank all the participants, especially the speakers from the different divisions, the FDA divisions, the speakers from the NCTR, and we'll wrap it up here, and we're going to continue tomorrow morning, 8 o'clock local time, NCTR, 9 o'clock on the east coast, so I think we'll follow the same procedure that we did today.

What I'd like to ask the participants to do -- not the participants, the other ones; are we the participants?

(Laughter.)

I'd like to ask the presenters to log in maybe 20 minutes, 25 minutes before 9 o'clock Eastern time, now we have some experience, so hopefully tomorrow will be a little bit faster.

So thank you very much. Have a good night.

Donna, I don't know if you want to add anything.

DR. MENDRICK: That is fine. Thank you so much, Miki, for all the help today and I certainly look forward to working with you tomorrow.

DR. ASCHNER: Thank you. We look forward to the meeting tomorrow. We will have three more presentations and then just a closed session for the SAB.

Good night and see you all tomorrow.

MICHAELKAWCZYNSKI: With that, today's meeting has ended. Go forth and have a great evening.

(Whereupon, the meeting was adjourned at 5 p.m.)